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Review

Potential causes of postharvest losses, low-cost cooling technology for fresh produce farmers in Sub-Sahara Africa

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The aim of this review was to identify the causes of postharvest losses (PHL) in fruit and vegetables in relation to small-scale farming in sub-Saharan Africa (SSA). The reduction of PHL can improve food security at household level. Farmers involved in small-scale production of fresh produce experience high PHL due to physiological deterioration associated with technical, biological and environmental factors and lack access to postharvest facilities. When these factors are contained, sufficient supplies of fresh produce reach the consumer and improve nutrition, income and food security at household level. This article described the PHL experienced by farmers along the cold chain and explored the advantages and disadvantages of the use of various cooling technologies. There are already existing modern cooling technologies but these are capital intensive and require electricity, which is not always available to small-scale farmers (SSF). This review proposes evaporative cooling as appropriate for SSF in SSA as it has proven to be effective under hot and dry areas and is a simpler and cheaper technology. The review recommends that with the incorporation of a desiccating unit, evaporative cooling could be extended to hot and humid areas. Solar and wind energy can be used to power the desiccating unit in remote and isolated areas with no access to grid electricity. Therefore, research needs to be carried out on developing or adapting a solar or wind powered evaporative cooling system under both hot-dry and hot-humid conditions.

Key words: Fruit and vegetables, low-cost cooling, postharvest technology, renewable energy, small-scale farming.

INTRODUCTION

Sub-Saharan Africa (SSA) has potential for fruit and vegetables (FV) production as there has been annual increases in price and quantities produced in the last five to ten years (Sibanda, 2019). Two distinct farming production levels, large-scale commercial agriculture and small-scale farming characterize the horticultural sector in SSA. In large-scale commercial farming, farmers own large tracts of land and have the financial capability to

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Table 1. Vegetable production per (1000t) and their average prices at major fresh produce market for 2010 and 2015 (DAFF, 2016).

Parameter	Vegetables production (1000t)		Average price at major fresh produce market (R/t)	
	2010	2015	2010	2015
Potatoes	1 955	2 423	2 598	3 222
Tomatoes	575	539	4 233	8 310
Pumpkins	234	256	1 737	1 805
Green mealies	339	373	8 260	13 726
Onions	489	675	2 573	2 802
Sweet potatoes	60	63	1 977	3 699
Green peas	17	9	17 960	37 012
Beetroot	67	78	2 763	3 050
Cauliflower	25	13	3 777	7 752
Cabbage	141	146	2 573	1 963
Carrots	151	201	3 251	2 132
Green beans	23	25	5 634	1 917
Lettuce	-	-	3 338	5 950

et al., 2012). Small-scale farmers (SSF) on the other hand on average own small land holdings of less than 1.5 ha and are characterized by low output and very little investment in infrastructure for production (Tscharntke et al., 2015). Despite these setbacks, SSF contribute approximately 80% of all FV farming activities in SSA (OECD/FAO, 2016).

The increasing population and shifts in consumer demand have resulted in an exponential demand and price hikes for fresh FV in SSA (Ntombela, 2012). For example, the demand has seen annual price increases in horticulture of 7% in South Africa (SAYB, 2015) and generally increased fresh produce quantities from 2010 to 2015 as shown in Table 1. This scenario should improve farmers' living conditions including health and income while at the same time ensuring food security at household level. An increasing demand for fresh produce at the right prices is likely to move SSF from subsistence to commercial scale farming. The greatest challenge constraining rural households from attaining commercial farming status is the quality deterioration experienced in the production cycle of fresh produce (Nkolisa et al., 2018). The quality of fresh produce can be maintained through provision of optimum storage conditions, which varies with crop type and depends on intended use, the level of quality required for the purpose, distance and time to market. However, in SSA appropriate post-harvest technologies for SSF have not been developed or adapted for the proper handling of perishable commodities (Cherono et al., 2018). With no appropriate postharvest facilities, which may include packaging, cooling technologies during storage and transportation, food security is threatened. The traditional peddling of fresh produce at farm gate at low prices to avoid losses is not an enduring solution as it ultimately undermines sustenance (Saran et al., 2012; Cherono and Workneh, 2019).

Furthermore, the fact that most SSF are located in remote areas with no access to grid electricity and have poor road infrastructure connecting them to major towns, hinders growth and productivity in small-scale farming (Kim and Ferreira, 2008; Korir et al., 2017). Small-scale farmers are in many instances forced to sale their produce to intermediaries (middle-men) that offer them low prices rendering their enterprises unprofitable (Cherono and Workneh, 2019; Sibanda, 2019).

Although there are a number of modern cooling technologies developed and imported into the region, SSF have not been able to adapt and utilise such facilities as they are both capital and energy intensive (Ejeta, 2009; Nkolisa et al., 2019). Despite the numerous researches on both production and postharvest handling of commodities in the region, there is less adaption or application of the research results to solve the post-harvest handling problems under SSA conditions particularly for small-scale farming (Stathers, 2017). Therefore, in order to discuss appropriate low cost cooling technologies this review has found it necessary to explore causes mainly related to postharvest physiology of crops since cooling mainly applies to slowing down respiration and ethylene production and the extent of postharvest losses (PHL). The review further considers different types of cooling technologies and explores alternative renewable energy options available for possible integration with low-cost technologies to preserve FV accessible by SSF.

Post-harvest losses

Postharvest losses are the quantifiable depression in a given produce during harvest or along the value chain of

Table 2. PHL in selected countries in SSA.

Sub-region	Country	Estimated postharvest losses (%)	References
East Africa	Ethiopia	50	FAO (2005)
Central Africa	Rwanda	30-80 depending on product	Kitinoja et al. (2010)
West Africa	Ghana	30-80 depending on product	Kitinoja et al. (2010)
Southern Africa	Swaziland	20-50	Masarirambi et al. (2010)

a post-harvest system (Sawicka, 2019). Fruit and vegetables are perishable commodities and highly susceptible to physiological deterioration in the supply chain, which is the primary reason of high PHL experienced in their production (Pathare et al., 2012; Singh and Sharma, 2018). Azene et al. (2011) claim that PHL have the potential to discourage farmers from venturing into production and marketing of fresh produce and thus affecting the availability and consumption of FV in mostly urban areas. Therefore, efforts that reduce PHL, particularly if they are economically feasible are of great significance to farmers and consumers alike (Miller et al., 2017). Reducing PHL, as an important component of food security, has potential to lower food prices to vulnerable communities in the region. In this food-scarce part of the world, FV not reaching the intended market are a significant waste of resources. In a survey carried out by Mashau et al. (2012) in the Tshakuma fruit market, in Limpopo province of South Africa showed that fresh fruits like bananas, oranges, avocados, paw-paws and tomatoes, experience deterioration in both quality and quantity of 43.3% mainly due to over-ripening.

Postharvest losses in the supply chain of fresh produce in SSA, are difficult to estimate as there is limited official data from different countries and there is no standard methodology to estimate the losses (Adeoye et al., 2009; Sibomana et al., 2016; Singh and Sharma, 2018). Postharvest losses in FV in the region estimate to over 50% though they vary from crop to crop and country to country (FAO, 2008; Kader, 2010; Mashau et al., 2012). Table 2 provides examples of estimated percentage PHL of perishable commodities for selected countries in East Africa, Central Africa, West Africa and Southern Africa. These high losses as shown in the Table 2 are a precursor to food insecurity for SSA communities. Small scale farming exporters of FV in the region have complained about these huge losses experienced during short periods of storage (awaiting transportation) and during transportation to markets and alleviation of these should be a research priority (Tigist et al., 2011).

Causes of PHL

Maintenance of fresh produce quality requires precise application of optimum cold chain conditions from harvest, grading, packaging, storage and transportation to the consumer and this review discusses these conditions. Postharvest losses occur due to many factors that include, environmental (Rayaguru et al., 2010), biological and chemical, physiological (Joas and Lechaudel, 2008), as well as technical factors (Kader, 2010). The main environmental factors that result in significant PHL in FV are temperature and relative humidity (Prusky, 2011; Bradford et al., 2018). The biological and chemical factors arise because FV are prone to microbial contamination during growth, harvest and postharvest operations (Ambaw et al., 2013a). Physiological deterioration of fresh produce happens since FV are living tissues (Sitorus et al., 2018). So, as lively tissues the produce continues to transpire, respire and further ripen even after they have been detached from the mother plant during harvesting (Ngcobo et al., 2012; Gupta and Dubey, 2018). As the anaerobic process continues, it in turn increases respiration further with even more heat generation either inside or outside the fruit. This sustained respiration in fresh produce means decreased food value, associated with loss of flavour, loss of saleable weight (through loss of moisture), and more rapid deterioration (Ait-Oubahou, 2013). The technical factors that affect fresh produce quality are mainly associated with mechanical damage or injury to FV (Paull and Duarte, 2011), lack of skilled labour in handling of fresh commodities (Beckles, 2012) and prolonged storage time (Wilson et al., 1999).

Controlling these factors provides improved efficiency of broader value chains and systems in fresh produce (Sawicka, 2019). On the other hand, social factors are associated with trends such as urbanization, where many people from rural areas move to large cities causing a high demand for FV at urban centres, thus increasing the need for more efficient supply-chains (Parfitt et al., 2010). The critical issue in all this is that the effects of the mentioned factors are not receiving the required attention at various control points such as harvesting, packaging, on-farm temporary storage and transportation to the market resulting in high PHL in the fresh produce supply chain.

Losses during harvesting and packaging

Harvest-labour should be skilled to know when to harvest the produce, as it is an essential requirement of industrial postharvest handling (Beckles, 2012). According to Bachmann and Earles (2014), harvesting of fresh produce should take place during the coolest part of the day, either early in the morning or in late afternoon. Harvesters also should be trained on how to handle the crop carefully to avoid injury; harvesting dry whenever possible and at proper maturity; handling each produce no more than is necessary, while at the same time avoiding careless handling e.g. dropping fresh produce (Prusky, 2011). Therefore, farmers have to exercise good harvesting practices that will not result in mechanical injury to fresh produce.

Van Zeebroeck et al. (2007) described mechanical damage as pausing a challenge to the quality of fresh produce and having a potential to reduce the value of FV. According to Basediya et al. (2011), mechanical injury due to impact when produce is dropped/tossed during harvesting can result in splitting of fruit and internal bruising. Impact damage is detrimental and its effect is not just limited to visual aspects but can also cause a risk of fungal and bacterial contamination, which may lead to a shorter shelf life (Tijskens, 2007). Mechanical damage to FV may result from inappropriate packaging or containers and over or under packaging of containers (Vigneault et al., 2009; Mashau et al., 2012). Packaging should ensure that produce is loaded into convenient units for handling during distribution, storage and marketing (Wills et al., 2007). However, many SSF in production of tomatoes utilise traditional baskets as packaging material (Ugonna et al., 2015). Whenever fresh produce is loaded in baskets/plastic crates, it applies a static load on itself (Adeoye et al., 2009). The static load results in excessive pressure applied in the lower part of the packaging material thus causing deformation of the produce at the bottom, which may result in bruising and breakage (Ugonna et al., 2015). This scenario occurs when baskets are used or when there is over-packaging. In under-packaging, the movement of fresh produce in the container is high, resulting in collision that damages the fruit (Prusky, 2011). In some instances, these plastic crates have rough internal surfaces, which can injure FV by contact (Sibomana et al., 2016).

Another cause of losses during harvesting and packaging is due to physiological deterioration of fresh produce as they continue respiration and ripening. The respiration rate of a product strongly determines its transit and postharvest life (Yahia, 2011). The higher the temperature at harvest, the higher the respiration rate will be, resulting in reduced shelf life (Sandhya, 2010).

Causes of losses during on-farm storage and transportation

Fruit and vegetables in some instances are stored at the farm gate for extended periods until either transport to the market becomes available or local buyers/market purchase the produce for consumption or resale (Hardenburg et al., 1986). Often the transport and local markets are without temperature-controlled environmental conditions resulting in further deterioration of fresh produce (Kitinoja and Thompson, 2010). In circumstances where on-farm storage and transportation facilities are not kept at below optimum environmental conditions, the ripening of FV continues resulting in physiological deterioration as fruit rot organisms spread most rapidly at warm storage temperatures and low relative humidity (Maliwichi et al., 2014; Sibomana et al., 2017). Physiological, chemical and enzymatic changes are speeded up when fresh produce is subjected to high ambient temperature and low relative humidity during temporary storage and transportation at the back of trucks (Fadeyibi and Osunde, 2011; Chijioke, 2017).

In some instances, the ambient temperatures in SSA especially in tropical and sub-tropical climates can be 7-20°C higher than the recommended 15°C for tomatoes (Kitinoja and AlHassan, 2012; Tolesa and Workneh, 2017). These two environmental factors can result in a significant loss of nutritional value, decreased returns due to poor produce quality (wilting, shrivelling), loss of saleable weight and in many cases the whole fruit or vegetable is lost (Odesola and Onyebuchi, 2009). Respiration rate, metabolic processes and ethylene biosynthesis of some fruits increase with air/room temperature within a given range (Workneh, 2010). Respiration rates can double, triple or even quadruple with every increase in temperature (Zagory and Kader, 1988). Therefore, the storage of FV at low temperature immediately after harvesting will reduce the rate of decomposition and microbial spoilage (Workneh and Osthoff, 2010). Fresh produce shelf life can double by reducing temperature from 10 to 5°C (Sun and Zheng, 2006). Typically, the storage temperature of FV is 0 to 12°C and most tropical and subtropical fruits require high temperatures of 5 to 13°C according to (Paull and Duarte, 2011) and as shown in Table 3.

Relative humidity is another important aspect considered during storage and transportation of FV. Occurrence of high humidity during temporary storage and transportation of fresh produce reduces water loss, helping FV maintain weight, appearance, nutritional quality and flavour, while wilting, softening and juiciness are reduced (Laguerre et al., 2013). According to Cantwell et al. (2009), the recommended storage relative humidity for most horticultural crops is 70 to 95%. Table 3 provides a summary of recommended storage relative humidity for selected FV. Most fresh produce under small scale production are stored at relative humidity levels lower than recommended resulting in excessive moisture loss (Singh et al., 2014). Subsequently, the FV suffer wilting, shrivelling and dryness resulting from small moisture losses of 3-6% (Nunes et al., 2009; Okanlawon and Olorumisola, 2017). These changes affect marketability of produce or economic value especially if FV are sold by weight (Yahia, 2011). Usually weight loss

Table 3. Optimum storage temperature and relative humidity of selected vegetables (Adopted from Krishnakumar, 2002).

Product	Optimum Temperature (°C)	Optimum relative humidity (%)
Broccoli	0	90-95
Cabbage	0	98-100
Lettuce	0	90-100
Carrots	0	98-100
Tomatoes	13-15	≥ 85
Guava	5-10	90
Mango	13	85-95
Potatoes	5-16	90-95
Onions	1-2	65-70
Garlic	0	65-70
Banana (green)	13-14	90-95
Cucumber	10-13	95

from perishable commodities is high if surrounding air temperature, flesh moisture content and temperature is high. Thus, under poor postharvest management conditions of storage or in transit, perishable commodities lose excessively large weight (Workneh, 2010).

Among other key contributors to high PHL in fresh produce are demographic and socio-economic characteristics of small-scale FV producers. Small-scale farmers have to travel to cities to sale their fresh produce and due to lack of transport; they tend to keep FV over long periods at the farm gate awaiting transportation to markets (Kader, 2003). When the waiting period at the farm gate is prolonged, there is further mechanical damage to produce due to over handling (Knee and Miller, 2002). The damaged FV allow easy penetration of microbial population into the tissue increasing chances of decay and growth of microorganisms (Fadeyibi and Osunde, 2011). As packaged produce applies static load on itself the degree of deformation on FV will depend on the period the static load is applied (Sirisomboon et al., 2011). The longer the period, the greater the deformation and stress effected on the produce. The stress effected on the produce will also depend on the ripeness of produce, as it ripens the same static load will inflict more internal flesh damage (Sibomana et al., 2016). The injury to produce increases if it is loaded at the back of trucks in rough road conditions because of vibration forces experienced (Kereth et al., 2013). For SSF in SSA, trucks that pick up produce are not regular and if a farmer misses the truck on a certain day it can take up to a week before there is transport to pick up his FV to the market (Sibanda, 2019). To eliminate this challenge, it is required that the duration between harvest and arrival at the markets be minimized (Sibomana et al., 2017)

Research in cold chain technologies and their costs and benefits

The maintenance of market quality of fresh produce through management of a cold chain is key to the success of the horticultural industry, it is therefore, not only necessary to cool down the product but to do so as quickly as possible after harvest (Paull, 1999). A cold chain is a temperature-controlled supply chain consisting of uninterrupted range of systems that monitor or maintain produce at a given temperature and keeps history (Aung and Chang, 2013). According to Prusky (2011), the requirements for maintaining quality and safety of horticultural perishables through the supply chain from harvest to consumption are the same in developing and developed countries. It is clear, however, that in SSA the challenges to be addressed go beyond whether or the fact that no cooling technologies exist. For SSF producing FV other factors come into play like volume cooled per day, harvest temperature versus recommended storage temperature, capital and operating costs (Azene et al., 2011). To invest in modern cooling technologies, SSF have to consider the cost-benefit analysis as to whether there will be an increased financial benefit associated with the chosen technology (Ejeta, 2009). Availability of electricity is one of the critical factors to consider as an energy input to power cooling technologies (Kitinoja et al., 2011).

Possible areas of consideration should allow low energy cool storage facilities so that fresh produce reaches markets at recommended storage conditions (Chaudhari et al., 2015). Achieving this would ensure that both the supply of fresh produce and the shelf life would improve significantly in SSA. Kitinoja and Thompson (2010) have previously reviewed pre-cooling systems for small-scale producers. These authors and broader literature have described various methods for preservation of fresh FV immediately after harvest. These cooling methods include among others, mechanical refrigeration (James et al., 2009; James and James, 2011), hydro-cooling (ASHRAE, 2011; Ambaw et al., 2013b), evaporative cooling (Ambaw et al., 2013b) and vacuum cooling (Wang and Sun, 2001; Zheng and Sun, 2006).

Performance of various types of cooling technologies

Mechanical refrigeration, forced air cooling, vacuum cooling, hydro-cooling and evaporative cooling of fresh produce have previously been described in detail by reviews that include Brosnan and Sun (2001) and Thompson et al. (1998) who placed emphasis to the different performance parameters of various cooling methods. Mechanical refrigeration refers to the process where heat absorption takes place at one point and heat dispersion at the other (Moureh et al., 2009). This is achieved through circulation of a refrigerant through the system by a compressor picking heat through the evaporator inside the fresh produce space and dissipating it through the condenser on the outside (Zou et al., 2006; Hera et al., 2007a). The compressor can be powered through an electric motor. The refrigeration system is energy intensive as electricity power is consumed throughout the cold chain (Hera et al., 2007b). This in turn leads to high product cost since unit energy costs make part of the unit cost for production of a given produce (Swain et al., 2009). However, where there is a ready and cheaper supply of electricity, mechanical refrigeration is the most reliable cooling technology (Kitinoja and Thompson, 2010).

Hydro-cooling is a fast, uniform cooling process of removing field heat from freshly harvested FV by bathing in chilled water or running cold water over it (Gomez-Lopez, 2012). Since the produce will be at higher temperature immediately after harvest, the heat movement takes place from the produce to the water leading to cooling (Rennie et al., 2003). This process is an efficient way to remove heat as it uses water, which removes heat at least five times faster than air (Bachmann and Earles, 2014). The use of water serves as a means of cleaning at the same time. Hydro-cooling reduces, water loss as the product is bathed in water, the rates of microbiological and biochemical changes in order to prevent spoilage and maintain quality and increase shelf life (Gustavsson et al., 2011). However, hydro-cooling has limitations in that it is only appropriate for commodities that tolerate wetting like carrots, peaches, asparagus, cherries etc. and is not appropriate for berries, potatoes to be stored, sweet potatoes, bulb onions, garlic, or other commodities that cannot tolerate wetting (Bachmann and Earles, 2014).

Vacuum cooling is a rapid evaporative cooling method for porous and moist foods to meet the special cooling requirements (Zhang and Sun, 2006). In this case cooling obtains by evaporation of moisture from the surface and within the produce (Sun and Zheng, 2006). Evaporation is encouraged and made more efficient by reducing the pressure to the point where boiling of water takes place at low temperature. The difference between vacuum cooling and conventional refrigeration is that for the former, the effect is achieved by blowing cold air or other cold medium over the product, and the later describes direct transfer of heat from a produce (Rennie et al., 2003). Speed and efficiency are the two features of vacuum cooling, which are unsurpassed by any conventional cooling method, especially when cooling boxed or palletised products (Sun and Wang, 2004). The speed and efficiency of vacuum cooling relate to the ratio between the evaporation surface and the mass of produce (Prusky, 2011). Cooling time, in order of 30 minutes, ensures that strict cooling requirements for safety and quality of foods can be met (Brosnan and Sun, 2001). Vacuum cooling is ideally for any product, which has free water, and the product structure cannot be damaged by the removal of such water.

Evaporative cooling or humidification of surrounding air in FV storage involves the use of principles of moist air properties or psychometrics (Workneh, 2007; Shahzad et al., 2018). In this system, temperature drops considerably and humidity increases to the suitable level for short–term on farm storage or transportation of perishables (Jha and Kudas Aleskha, 2006). Evaporative cooling provides cool air by forcing hot dry air over a wetted pad (Chaudhari et al., 2015). The water in the pad evaporates, removing heat (sensible heat) from the air while adding moisture. Evaporative cooling is regarded as a low-cost cooling system requiring no electricity input in a passive system or just an electric fan in a forced air system (Tigist et al., 2011; Chijioke, 2017; Sibanda and Workneh, 2019). Evaporative cooling has been reported for achieving a favourable environment in storage structures for FV where shelf life of some fresh produce like apples, tomatoes, bananas, mangoes, potatoes and pumpkins has been increased by factors of 1.3-5 at the same time exhibiting good appearance (Chaudhari et al., 2015). In the work done by Anyanwu (2004) evaporative cooling increased, the shelf life of tomatoes by a factor of three above open-air storage values.

Modern cooling technologies like, mechanical refrigeration, vacuum cooling and hydro-cooling could be used in SSA depending on, the type of fresh produce, the rate of cooling required, energy consumption requirements, level of production, availability of funds to purchase the technology and availability of energy. Regrettably, most SSF in SSA are located in areas where there is no grid electricity for driving these modern cooling technologies. There are also issues related to, the cost of modern cooling technologies, performance of modern cooling technologies, economies of scale and relevance to small-scale production under SSA conditions discussed in the next section.

Selection of a suitable cooling technology for different fruits and vegetables

Where there is, uninterrupted electricity supply, investment capital is not limited to cover purchase and

Table 4. Summary of advantages, disadvantage and characteristics of different cooling technologies

Cooling technology	Advantages	Disadvantages	Performance characteristics	References
Evaporative cooling	Low capital cost; high energy efficient; environmental benign; low weight loss; slow deterioration in quality; suitable for rural application; requires no special skill to operate; can be made from locally available materials; and easy to maintain.	Requires a constant water supply; no humidification, and high dew point; condition decreases the cooling capability; mineral deposits leading to pad and interior damage	Can maintain temperatures at 10-15°C below ambient; Can achieve relative humidity of 90%; Can increase shelf life from 3 days to 15 days. Typical cooling time is 40-100 h in passive cooling and 20-100 hours in fan-ventilated systems.	Mordi and Olorunda (2003) Anyanwu (2004) Basediya et al. (2011) Chaudhari et al. (2015) Tigist et al. (2011)
Hydro-cooling	Rapid cooling; prevents loss of moisture during cooling; cools and cleans the produce at the same time; and simple and effective pre-cooling method; High energy efficient.	Not uniform may leave "hot spots"; not suitable for leafy produce; not suitable for products that do not tolerate wetting; not suitable for products that can be damaged by falling water; water left on surface can lead to fungus growth or discoloration; capital cost is relatively high; the equipment is not portable	Cooling can be achieved in 20-30 min; Water removes heat about 15 times faster than air at typical flow rates and temperature difference; Refrigeration capacity of 1.4 kW cool 500 kg produce per hour to achieve 11°C depression;	Wills et al. (2007) Brosnan and Sun (2001) Rennie et al. (2003) Prusky (2011)
Vacuum cooling	Rapid cooling achievable; distinct advantage over other cooling methods; cooling can achieve uniform cooling; gives highest energy efficiency; and hygienic since air only goes to the vacuum chamber; No potential for decay contamination; equipment is portable.	Very capital cost; limited application to large growers; causes weight loss in the produce; only suited for produce with a high surface to volume ratio; works best only for produce like lettuce; cabbage, mushroom	Rapid cooling; method and can achieve temperatures of 1°C; Can increase shelf life from 3-5 days at ambient temperature to 14 days when combined with cold storage at 1°C; For every 5.5°C reduction in temperature there is 1% weight loss;	Turk and Celik (1993) Kim et al. (1995) Ito et al. (1998) Brosnan and Sun (2001) Rennie et al., (2003)

cost of installation, availability of technical skills to maintain and run the facility, mechanical refrigeration would be the ideal cooling system (Basediya et al., 2011). However, mechanical refrigeration is not suitable for several FV; for example, banana, plantain, tomato etc. cannot be stored in the domestic refrigerator for a long period as such produce is susceptible to chilling injury (Chinenye, 2011). A small scale commercial mechanical refrigeration system with a capacity of one tonne complete and ready for use in the USA will cost about US$7000 for 3.5 kW (Kitinoja and Thompson, 2010). This cost is way above what most SSF in the region can afford for a cooling capacity of one tonne. Table 4 describes the advantages; disadvantages and the performance characteristics of evaporative cooling, vacuum cooling and hydro-cooling. The selection of suitability of each cooling technology for a certain crop will depend on such performance characteristics and parameters.

Hydro-cooling, is only suitable for leafy produce and has other limitations of low energy efficiency, requirement of expensive water resistant containers to avoid cross decay contamination (Thompson et al., 1998; Vigneault et al., 2000). The application of hydro-cooling by SSF will be limited by its unsuitability to cooling of root and grass crops and vegetables like tomatoes, apples and pepper as they have a thick cuticle (Wang and Sun, 2001). Vacuum cooling is only suitable for fresh produce with a high ratio of surface to volume and is unsuitable for oranges, tomatoes and apples (McDonald and Sun, 2000). Any cooling method unsuitable for tomatoes would be unattractive as this fruit is a major commodity grown for SSF in a number of countries in the region (Mashau et al., 2012).

Both vacuum cooling and hydro-cooling are regarded as energy intensive and expensive methods for example it would require for hydro-cooling, 110 to 150 kWh (15-22 kWh for vacuum cooling) of energy at a cost US$22-US$30 to cool one metric tonne of fresh produce (Kitinoja and Thompson, 2010; Rayaguru et al., 2010; Basediya et al., 2011). Vacuum cooling and hydro-cooling therefore, need to be operated for relatively longer periods in a year to justify an investment (Boyette et al., 1994). Brosnan and Sun (2001) concluded that since the vacuum chamber system for vacuum cooling was expensive then this cooling technology was only feasible for large growers who produce large volumes of fresh produce throughout the year. Unfortunately, SSF in SSA do not have sufficient volumes of fresh produce to warrant the use of vacuum and hydro cooling throughout the year (Kitinoja et al., 2011). As a result, these two cooling methods are limited for products for which they are much faster and more convenient like cherries for hydro-cooling and lettuce, cabbage, mushroom or produce with high surface to volume ratio for vacuum cooling (Kim et al., 1995; Thompson et al., 1998; Brosnan and Sun, 2001; Kitinoja and Thompson, 2010).

Another limiting factor of the use of hydro-cooling and vacuum cooling by SSF is that both are pre-cooling methods, refrigeration is still required thereafter between the farm and the market. The construction and operating costs of different cooling technologies vary from relatively low to high depending on the level of farm management (Kitinoja et al., 2011). Sometimes farmers would often ignore the cost of cooling technique during selection of technology as they transfer the cost to consumers making selling price of the produce higher especially in developed countries where there is a good marketing system (Boyette et al., 1994). In developing countries where intermediaries set prices at farm gate, SSF may find themselves selling their produce below the production costs (Cherono and Workneh, 2018).

Evaporative cooling could provide a solution, as the cooling technology has low initial investment, low installation and maintenance costs and in a passive system can be set up without electricity (Nkolisa et al., 2019). It is possible to construct an evaporative cooling system of 1-2 MT at US$1,300 at an energy use per MT of 0.7 kWh (Kitinoja and Thompson, 2010). The energy costs to cool one MT of tropical FV using evaporative cooling is $0.14 (Kitinoja and Thompson, 2010). Evaporative cooling presents itself as an appropriate cooling technology for small-scale farming of fresh produce in SSA as it is suitable for sub-tropical and tropical FV, the volumes for cooling per farmer per unit time are not huge, the storage temperature is around 15°C. Chaudhari et al. (2015) reviewed the work done on evaporative cooling from 1987 to 2010 and concluded that this system is not harmful to the environment, has low initial costs, can be constructed from local available material (storage chamber, cooling chamber, water tank, cooling pad media). Components that require maintenance like the motor, extraction fan and heat exchanger are repairable at low cost (Deoraj et al., 2015) and therefore, what is only left is finding relevant and cheap energy sources for its up scaling.

Relevance of evaporative cooling to small-scale farmers in SSA

A number of studies have shown the attractiveness in the use of evaporative coolers by SSF in Africa as evidenced by the work of a number of authors; Anyanwu (2004) in Nigeria, Ahmed et al. (2011) in Sudan, Samira et al. (2011) in Ethiopia. The results of use of evaporative cooling have demonstrated that coolers can maintain cooling spaces at temperatures below ambient with a depression reaching 12°C (Anyanwu, 2004). In evaporative cooling, lies the solution for SSF in finding a method appropriate that could alleviate storage challenges, reduce losses and improve food security at household level (Mordi and Olorunda, 2003). Should a forced air systems be required through use of a fan, the energy requirements are low and the cooling technology is energy efficient and a possibility exists to integrate it with use of alternative energy like wind or solar energy (Sibanda and Workneh, 2020). Fossil fuels could power the cooling methods but these contribute to greenhouse gas emissions (Best et al., 2012).

RENEWABLE ENERGY USE IN POSTHARVEST HANDLING

Renewable energy technologies have a high adaptation rate in many industries due to benefits related to climate mitigation, ability to enter foreign markets because of green processes, green consumer requirements and improved corporate images of industries that use clean energy (OECD/IEA and IRENA, 2017). Besides conventional energy sources there is an option of energy provision from natural energy sources that include among others solar and wind energy (Mentis et al., 2015). The consideration of the role of renewable energy along the different stages of food supply chain by providing requisite energy supplies especially for powering the fresh produce cold chain is important (Chaudhari et al., 2015). This is especially true for remote, dispersed populations with low and scattered energy demands. Both solar and wind energy represents the largest source of renewable energy supply compared to solid biomass, biogas, hydro and geothermal sources (Tyagi et al., 2012).

The consumption of fossil fuel is the major contributor to the greenhouse gases emitted to the atmosphere thus causing global warming (Hassan and Mohamad, 2012). Biomass is combusted for heating and cooking and is convertible into electricity (David et al., 2002). Direct combustion of biomass produces steam, which turns turbines that drive generators, producing electricity (Ayhan, 2006). The cost of producing 1 kW of electricity from wood biomass is US$0.058. Biomass combustion releases different chemical pollutants, including fourteen carcinogens into the atmosphere (Godish, 1991). Grid electrification is expensive and yet other sources of energy can meet all the energy requirements (Deveci et al., 2015). Senol (2012) recognizes the need to promote alternative energy supply especially for increased productivity and for income generation.

Wind energy or power is the production of electricity by turning blades on a wind turbine (Ayhan 2006). The advantage of wind turbines over other renewable energy sources is that they can produce electricity whenever the wind blows (both during the day and at night). Wind energy can be utilised if the annual energy available is at an average speed of 5 ms^{-1}, and is 490 $MJ.m^{-2}$ of surface perpendicular to the wind flux (Mentis, 2013). According to Archer and Jacobson (2005) and Mentis et al. (2015), while Africa has an abundance of wind energy, in some areas it is seasonal while in coastal regions is available throughout the year.

Solar energy seems to be the most viable alternative to fossil fuels as it is clean and renewable since it comes from the sun (Sontake and Kalamkar, 2016). Solar energy is the largest source of renewable energy supply, compared to solid biomass, biogas, hydro, wind etc. and is available in most areas of SSA throughout the year with values in excess of 2000 kWh m^{-2} (Davis and MacKay, 2013). In this region, the average solar radiation ranges between 4.5 to 6.5 $kWh.m^{-2}$ for an average of 6 -7 h (Fluri, 2009). This, according to Saxena et al. (2013), is enough solar radiation that is convertible to electricity. There has been application of solar energy in generating solar thermal or directly conversion to electricity through photovoltaic cells (Hassan and Mohamad, 2012). There is a lot of research work currently for absorption based refrigeration and air conditioning systems that use solar energy (Said et al., 2012). Solar energy has also been integrated with evaporative cooling by many researchers (Tiwari and Jain, 2001; Maerefat and Haghighi, 2010; Naticchia et al., 2010) for cooling of buildings. Naticchia et al. (2010) exploited both air ventilation and heat exchange by use of porous insulating material as an absorption matrix. Maerefat and Haghighi (2010) integrated a solar system employing a solar chimney with evaporative cooling cavity. This integrated system enhanced passive cooling and natural ventilation in a solar house, and the numerical experiments showed that daytime temperatures significantly reduced at a poor solar intensity of 200 $W.m^{-2}$ and high ambient temperature of 40°C. Finocchiaro et al. (2012) employed a solar energy assisted desiccant and evaporative cooling system for building air conditioning. In this system, solar energy regenerated a desiccant material that dehumidifies moist air by vapour adsorption. The resultant dry and warm air was then cooled in a sensible heat exchange and then in an evaporative cooler.

Because of research work, there have been reasons for focusing on the potential of converting solar energy through photovoltaic systems for use in agriculture production. This could be a basis for sustainable agricultural production at village level in SSA. The challenge is for researchers to find means of dramatically reducing the cost per solar panel to deliver cheaper energy to SSF. It is believed that this has been achieved to a certain extent as the price of renewable energy from solar has dropped in the last decade from US$0.18 kWh to just US$0.03 kWh (OECD/IEA and IRENA 2017).

Relevance of solar energy in evaporative cooling of fresh produce

Best et al. (2012) estimate that energy demand for cooling processes and greenhouse gas emissions will increase by 60% by 2030 compared to 2000 levels. Kim and Ferriera (2008) have recognised that there are energy requirements for agriculture in rural areas addressed by using alternative sources of energy other than grid electricity. Efforts in planning and provision of the additional power requirements with clean energy need to be in place. In Africa, there are more opportunities to use renewable energy because much of the continent has limited access to grid electricity (Szabo et al., 2011).

Therefore, the high-energy demands on existing power sources and global warming threats provide impetus for research towards technological alternatives (Hassan and Mohamad, 2012). Among these technologies, solar energy is the most appropriate for adaptation with cooling methods for fresh produce, as the resource is available throughout the year (Best et al., 2012). A lot of research in this regard has been taking place. The use of solar energy for evaporative cooling in all the cases has been limited to buildings and this provides an opportunity for the extension of the same principles to the preservation of fresh produce. The use of solar energy to power a water pump and fan is very limited and literature was not found providing evidence that wind energy has been used for evaporative cooling for fresh produce.

Evaporative cooling technology if used with forced air requires lower energy to operate water pump and fans while it is effective in providing cold and humid air to the storage chamber. The use of photovoltaic solar energy to operate low-cost cooling technologies for FV has a high potential. However, engineering design especially to convert solar energy into electrical energy and the storage of this energy in a battery to run the technologies during night remains one of the important research and development areas that need the attention of engineers. Similarly, wind energy is also suitable to provide sufficient energy to operate simple low-cost cooling technology that is appropriate for temporary storage of FV by SSF. Hence, an integrated approach of evaporative cooling and renewable energy as a source of power could be highly suitable for SSF that are engaged in production of FV in SSA. This will play a pivotal role in ensuring food security at household level and a reliable family sustenance through income obtained from sales.

DISCUSSION

All categories of farmers' experience high PHL in SSA. The deterioration in quality of FV is largely due to factors such as technical, biological and chemical, and as well as environmental aspects. These factors affect fresh produce quality from harvesting, packaging, temporary storage at the farm through to transportation to markets. Literature shows that the introduction of appropriate cooling technologies for SSF will ensure provision of cold chain systems that minimize PHL from harvesting to consumption by end user of fresh produce. The training of harvesters and ensuring the use of appropriate transportation containers are important to reduce the effect of technical factors on PHL. It is evident from literature that biological processes play a key role in aggravating PHL if not properly controlled. The control of temperature and relative humidity is through the introduction of appropriate cold chain storage facilities in the produce supply chain. This review identified a number of modern cooling technologies available in the market such as vacuum cooling, hydro-cooling and mechanical refrigeration. The different modern cooling technologies have inherent challenges in their application by SSF in SSA as they are energy intensive, expensive, require sustained higher volumes throughout the year and their use is specific to certain types of FV. However, this review showed that in developing countries like in SSA there is lack of access to proper cold chain storage facilities because of these aforementioned challenges.

Hence, there is a need to identify, develop or adapt appropriate low cost cold chain facilities for access by SSF. This is the only way SSF can rise from subsistence farming to commercial fresh produce production. Further, this review also recognizes that evaporative cooling is a simple and cheap method compared to conventional cooling technologies. Evaporative cooling also premises on removal of sensible heat, which makes it relatively efficient under hot and dry environmental conditions obtained in SSA. Evaporative cooling does not necessarily need an external power source as it relies on velocity of natural wind through wetted pads. Evaporative cooling is ideal, for both pre-cooling and cooling and observations are that it increases shelf life of fresh produce. Evaporative cooling has had a big impact in cooling of buildings in Asia and is practiced by some SSF in SSA. At the same time, many SSF are practicing natural ventilated evaporative cooling as they do not have access to grid electricity to power forced air evaporative cooling.

Grid electricity is not available in remote and isolated areas in SSA, while fossil fuels as energy source have a limitation in that they emit greenhouse gases. The immediate alternative then is the use of renewable energy sources like solar and wind, which are abundant in SSA. As a result, there exists a research scope in the utilisation of wind and solar energy to support evaporative cooling of fresh produce. This integrated system could be very useful to SSF in SSA producing FV in ensuring that they rise from high PHL incurring farmers to profitable farmers who are able obtain returns enough to sustain their families. Therefore a scope exists develop an integrated system that involves renewable energy sources like solar or wind combined with a cooling technology. There is no evidence of research work on the use of wind energy as a power source for cooling technologies. However, there is potential in the use of solar energy to power non-passive evaporative cooling for buildings. These success stories in this regard provide an opportunity for the use of solar energy to power a water pump and fan to drive non-passive evaporative cooling. From the conclusions made above, the proposition is carrying out a study to develop or adapt a wind and solar powered evaporative cooling system for temporary storage and transportation of FV.

RECOMMENDATIONS

Literature has shown that evaporative cooling is effective in hot and dry areas but has limitations in hot and sub-humid to humid areas because of inherent high humidity of the local air, which leads to low dry bulb temperature drops. It is recommended that researchers incorporate a desiccating unit for sensible cooling of air before reaching the cooling pads for evaporative cooling to find expression in hot and humid areas. Therefore, from the review, researchers can consider to develop and evaluate a solar or wind energy powered evaporative cooling system combined with an indirect air-cooling unit for use in hot and humid areas for storage of fresh produce.

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CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Adeoye B, Odeleye O, Babalolaand O, Afolayan O (2009). Economic analysis of tomato losses in Ibadan Metropolis, Oyo estate, Nigeria. African Journal of Basic and Applied Science 1(5-6):87-92.

Ahmed M, Abaas O, Ahmed M, Ismail R (2011). Performance evaluation of three types of local evaporative cooling pads in greenhouses in Sudan. Sudanese Journal of Biological Science 18:45-51.

Ait-Oubahou A (2013). Postharvest technologies in sub-Saharan Africa: status, problems and recommendations for improvements. In Acta Hortic; International Society of Horticultural Science (ISHS): Leuven, Belgium, pp. 1273-1282.

Ambaw A, Delele A, Defraeye T, Ho T, Opara U, Nicolai M, Verboven P (2013a). The use of CFD to characterize and design post-harvest storage facilities: Past, present and future. Computers and Electronics in Agriculture 93:184-194.

Ambaw A, Verboven P, Defraeye T, Tijskens E, Schenk A, Opara UL, Nicolai BM (2013b). Effect of box materials on the distribution of 1-MCP gas during cold storage: A CFD study. Journal of Food Engineering 119:150-158.

Anyanwu EE (2004). Design and measured performance of a porous evaporative cooler for preservation of fruit and vegetables. Energy Conversion and Management 45:2187-2195.

Archer CL, Jacobson MZ (2005). Evaluation of global wind power. Journal of Geophysical Research 120(12):1-20.

ASHRAE (2011). Standard for the design of high-performance green buildings. American Society of Heating, Refrigerating and Air Conditioning Engineers Inc., Atlanta, GA. ASHRAE/USGBC/IES standard 189:1-2011.

Aung MM, Chang Y (2013). Temperature management for the quality assurance of a perishable food supply chain. Food Control 40(1). Available at: https://doi.10.1016/j.foodcont.2013.11.016.

Ayhan D (2006). Global Renewable energy resources, Energy Sources, Part A: Recovery, Utilization, and environmental effects 28(8):779-792. Available at: https://doi.org.10.1080/00908310600718742.

Azene W, Workneh TS, Woldestadik K (2011). Effect of packaging materials and storage environment on postharvest quality of papaya fruit. Journal of Food Science and Technology. Available at: https://doi.org.10.1007/s13197-011-0607-6.

Bachmann J, Earles R (2014). Postharvest of fruit and vegetables. (Internet). National Center for Appropriate Technology (NCAT). Available at: https://attra.ncat.org/attra-pub/viewhtml.php?id=378. [Accessed 08 January 2020].

Basediya ALD, Samuel VK, Beera V (2011). Evaporative cooling system for storage of fruit and vegetables – a review. Food Science Technology. Available at: https://doi.org.1007/s13197-011-0311-6.

Beckles DM (2012). Factors affecting the postharvest soluble solids and sugar content of tomato (Solanum lycopersicum L.) fruit. Postharvest Biology and Technology 63:129-140.

Best B, Aceves JJ, Islas HJM, Manzini SFB, Pilatowsky PIF, Scoccia R, Motta M (2012). Solar cooling in the food industry in Mexico: A case study. Applied Thermal Engineering. Available at: https://doi.10.1016/j.applthermaleng.2011.12.036.

Boyette MD, Wilson LG, Estes EA (1994). Hydro-cooling. North Carolina Cooperative Extension Services, Raleigh /North Carolina Agricultural and Technical State University, Greensboro AG-414-4.

Bradford KJ, Dahal P, Asbrouck JV, Kunusoth K, Bello P, Thompson J, Wu F (2018). The dry chain: Reducing postharvest losses and improving food safety in humid climates. Trends in food Science and Technology 71:84-93. Available at: https://doi.org/10.1016/j.tifs.2017.11.002

Brosnan T, Sun D (2001). Precooling techniques and applications for horticultural products–a review. International Journal of Refrigeration 24(2):154-170.

Cantwell MI, Nie X, Hong G (2009). Impact of storage conditions on grape tomato quality. Sixth ISHS Postharvest Symposium, International Society of Horticultural Science, Antalya, Turkey.

Chaudhari, BC, Sonawane, TR, Patil SM, Dube A (2015). A review on evaporative cooling technology. International Journal of Research in Advent Technology 3(2):88-96.

Cherono K, Workneh TS (2018). A review of the role of transportation on the quality changes of fresh tomatoes and their management in South Africa and other emerging markets. International Food Research Journal 25(6):2211-2228.

Cherono K, Sibomana M Workneh TS (2018). Effect of infield handling conditions and time to pre-cooling on the shelf-life and quality of tomatoes. Brazilian Journal of Food Technology https://doi.org/10.1590/1981-6723.01617.

Chinenye NM (2011). Development of clay evaporative cooler for fruit and vegetables preservation. Agricultural Engineering International, CIGR Journal 13(1).

Chijioke OV (2017). Review of evaporative cooling systems. Greener Journal of Science, Engineering and Technological Research 7(1):1-20.

DAFF (2016). Abstracts of agricultural statistics. Department of Agriculture, Forestry and Fisheries. Republic of South Africa.

David P, Megan H, Michele G, Mathew Z, Richard A, Katrina B, Jeff E, Benita H Ryan S, Anat, G, Thomas S (2002). Renewable Energy: Current and Potential Issues. Biological Science 52(12):1111-1120. Available at: https://doi.org.10.1641/0006-3568(2002)052.

Davis J, MacKay F (2013). Solar Energy in the Context of Energy Use, Energy Transportation, and Energy Storage [Internet]. University of Cambridge. Cambridge, UK. Available from: http://www.inference.eng.cam.ac.uk/sustainable/book/tex/RSsolar.pdf [Accessed 17 April 2016].

Deoraj S, Ekwue EI, Birch R (2015). An evaporative cooler for storage of fresh fruits and vegetables. West Indian Journal of Engineering 38(1):86-95.

Deveci O, Onkol M, Unver HO, Ozturk Z (2015). Design and development of a low-cost solar powered drip irrigation system using systems modelling language. Journal of Cleaner Production 102:529-544.

Ejeta G (2009). Revitalising agricultural research for global food security. Food Security 1:391-401.

Fadeyibi A, Osunde ZD (2011). Measures against damage of some perishable products on transit. Advances in Agriculture, Sciences and Engineering Research 1(1):1-8

FAO (2005). Harvest handling and losses. Food and Agriculture Organisations of the United Nations, Rome.

FAO (2008). The world vegetable center. Newsletter. Food and Agriculture Organisations of the United Nations, Rome.

Finocchiaro P, Beccal, M, Nocke B (2012). Advanced solar assisted desiccant and evaporative cooling system with wet heat exchangers. Solar Energy 86:608-618.

Fluri TP (2009). The potential of concentrating solar power in South Africa. Energy Policy 37(12):5075-5080.

Godish T (1991). Air Quality. Chelsea (MI): Lewis Publishers.

Gomez-Lopez VM (2012). Decontamination of Fresh and Minimally Processed Produce. Oxford, United Kingdom: John Wiley and Sons. Inc., 9600 Garsington Riad.

Gupta J, Dubey RK (2018). Factors affecting post-harvest life of flower crops: A review. International Journal of Current Microbiology and Applied Sciences 7(1):548-557. Available at: https://doi.org/10.20546/ijcmas.2018.701.065.

Gustavsson J, Cederberg C, Sonesson U, Van Otterdijk R, Meybeck A (2011). Global food losses and food waste: extent, causes and prevention, FAO Rome.

Hardenburg RE, Watada AE, Wang CY (1986). The Commercial Storage of Fruits, Vegetables, and Florist and Nursery Stocks, USDA Handbook No. 66 (revised), 136. USDA, Washington, USA.

Hassan HZ, Mohamad AA (2012). A review on solar cold production through absorption technology. Renewable Energy and Sustainable Energy Reviews 16:5331-5348.

Hera D, Drughean L, Girip A (2007a). Improvement of the energy efficiency in the refrigeration plants. 22nd International Congress of Refrigeration 1:859-865. ISBN: 978-1-62276-045-9.

Hera D, Ilie A, Dumitrescu R (2007b). Aspects regarding the energy efficiency's increase in the case of refrigeration systems and heating pumps in Bucharest-Romania. 22nd International Congress of Refrigeration 5: 4136-4143. ISBN: 978-1-62276-045-9.

Ito H, Takase N, Sato E (1988). Effect of low temperature on keeping quality of butterbur during distribution. Research Bulletin of the Aichi ken Agricultural Research Center 20:269-277, ISSN 0388-7995.

James SJ, Swain MJ, Brown T, Evans JA, Tassou SA, Ge YT, Eames I, Missenden J, Maidment G, Baglee D (2009). Improving the energy efficiency of food refrigeration operations. Proceedings of the Institute of Refrigeration, Session 2008-09. 5-1-5-8.

James SJ, James C (2011). Improving energy efficiency within the food cold chain. 11th International Congress on Engineering and Food (ICEF) 22-26 May 2011. Athens, Greece.

Jha SN, Kudas Aleskha SK (2006). Determination of physical properties of pads for maximising cooling in evaporative cooled store. Agricultural Engineering 43(4):92-97.

Joas J, Lechaudel M (2008). A comprehensive integrated approach for more effective control of tropical fruit quality. Stewart Postharvest Review 4:1-14.

Kader AA (2003). A perspective on postharvest horticulture (1978-2003). HortScience 38:1004-1008.

Kader AA (2010). Handling horticultural perishables in developing countries versus developed countries. Acta Horticulturae 877:121-126.

Kereth GA, Lymo M, Mbwana HA, Mongi RJ, Ruhembe C (2013). Assessment of postharvest handling practices: knowledge and losses of fruits in Bagamoyo district of Tanzania. Food Science and Quality Management 11:8-15.

Kim BS, Kim DC, Lee SE, Nahmgoong, Choi MJ, Joong MC (1995). Freshness prolongation of crisp head lettuce by vacuum cooling. Agricultural Chemistry and Biotechnology 38(3):239-247.

Kim DS, Ferreira CAI (2008). Solar refrigeration options – a state of the art review. International Journal of Refrigeration 31:3-15.

Kitinoja L, AlHassan HA, Saran S, Roy SK (2010). Identification of appropriate postharvest technologies for improving market access and incomes for small horticultural farmers in sub-Saharan Africa and South Asia. IHC Postharvest Symposium August 23, 2010. Lisbon, Portugal.

Kitinoja L, Thompson JF (2010). Pre-cooling systems for small-scale producers. Stewart Postharvest Review. Available at: https://doi.org.10.2212/spr.2010.2.2.

Kitinoja L, Saran S, Roy SK, Kader AA (2011). Postharvest technology for developing countries: challenges and opportunities in research, outreach and advocacy. Journal of Science Food Agriculture 91:597-603.

Kitinoja L, AlHassan HY (2012). Identification of appropriate postharvest technologies for small-scale horticultural farmers and marketers in sub-Saharan Africa and South Asia – Part 1. PHL and quality assessments. Acta Horticulturae 31-40.

Knee M, Miller AR (2002). Mechanical Injury. In: (ed) Knee, M. Fruit quality and its biological basis. Sheffield academic press, Sheffield, pp. 157-179.

Korir MK, Mutwiwa UN, Kituu GM, Sila DN (2017). Effect of near infrared reflection and evaporative cooling on quality of mangoes. Agricultural Engineering International: CIGR Journal 19(1):162-168.

Krishnakumar T (2002). Design of cold storage for fruits and vegetables. Technical report. Available at: https://doi.org.10.131340/RG.2.2.14335.82082.

Laguerre O, Hoang HM, Flick D (2013). Experimental investigation and modelling in the food cold chain: Thermal and quality evolution. Trends in Food Science and Technology 29:87-97.

Maerefat M, Haghighi AP (2010). Natural cooling of stand-alone houses using solar chimney and evaporative cooling cavity. Renewable energy 35:2040-2052.

Maliwichi LL, Pfumayaramba TK, Katlego T (2014). An analysis of constraints that affect smallholder farmers in the production of tomatoes in Ga-Mphahlele, LepelleNkumbi municipality, Limpopo Province, South Africa. Journal of Human Ecology 47(3):269-274.

Masarirambi MT, Mavuso V, Songwe VD, Nkambule TP, Mhazo N (2010). Indigenous postharvest handling and processing of traditional vegetables in Swaziland: A review. African Journal of Agricultural Research 5(24):3333-3341.

Mashau ME, Moyane JN, Jideani IA (2012). Assessment of post-harvest losses of fruits at Tshakhuma fruit market in Limpopo province, South Africa. African Journal of Agricultural Research 7(29):4145-4150.

McDonald K, Sun DW (2000). Vacuum cooling technology for the food processing industry. Food Engineering 45:55-65.

Mentis D (2013). Wind Energy Assessment in Africa A GIS-based Approach. Unpublished Master of Science Thesis, KTH School of Industrial Engineering and Management, vetenskap och konst KTH, Stockholm, Sweden.

Mentis D, Hermann S, Howells M, Welsch M, Siyal SH (2015). Assessing the technical wind energy potential in Africa a GIS-based approach. Renewable Energy 83:110-125.

Miller V, Mente A, Dehghan M, Rangarajan S, Zhang X, Swaminathan S, Dagenais G, Gupta R, Mohan V, Lear S, Bangdiwala SI, Schutte AE, Wentzel-Viljoen E, Avezum A, Altuntas Y, Yusoff K, Ismail N, Peer N, Mapanga R (2017). Fruit, vegetable and legume intake, and cardiovascular disease and deaths in 18 countries (PURE): a prospective cohort study. The Lancet 390(10107):2037-2049. Available at: https://doi.org/10.1016/S0140-6736(17)32253-5.

Mordi JI, Olorunda AO (2003). Effect of evaporative cooler environment on the visual qualities and storage life of fresh tomatoes. Food Science Technology 40(6):587-591.

Moureh J, Tapsoba S, Derens E, Flick D (2009). Air velocity characteristics within vented pallets loaded in a refrigerated vehicle with and without air ducts. International Journal of Refrigeration 32(2):220-234.

Naticchia B, D'Orazio M, Persico I (2010). Energy performance evaluation of a novel evaporative cooling technique. Energy and buildings 42:1926-1938.

Ngcobo MEK, Delele MA, Opara UL, Zietsman CJ, Meyer CJ (2012). Resistance to airflow and cooling patterns through multi-scale packaging of table grapes. International Refrigeration 35(2):445-452.

Nkolisa N, Magwaza LS, Workneh TS, Chimphango A (2018). Postharvest quality and bioactive properties of tomatoes (Solanum lycopersicum L.) stored low-cost and energy-free evaporative cooling system. Science of Horticulture 241:131-143. Available at: https://doi.org//10.1016/j.scienta.2018.06.079.

Nkolisa N, Magwaza LS, Workneh TS, Chimphango A, Sithole NJ (2019). Evaluating evaporative cooling system as an energy-free and cost- effective method for postharvest storage of tomatoes (Solanum lycopersicum L.) for smallholder farmers. Heliyon Journal. Available at: https://doi.org/10.1016/j.heliyon.2019.e02266

Ntombela S (2012). South African fruit trade flow. Promoting market access for South African agriculture. Issue No. 6, June 2012. National Agricultural Marketing Council, Pretoria, South Africa.

Nunes MCN, Emond JP, Rauth M, Dea S, Chauk V (2009). Environmental conditions encountered during typical consumer retail display affect fruit and vegetable quality and waste. Postharvest Biology and Technology 51(2)232-241.

Odesola IF, Onyebuchi O (2009). A review of porous evaporative cooling for the preservation of fruit and vegetables. Pacific Journal Science Technology 27(1):19-21.

OECD/FAO (2016). Agriculture in sub-Saharan Africa: Prospects and challenges for the next decade in OECD-FAO Agricultural Outlook 2016-2025, Paris.

OECD/IEA and IRENA (2017). Chapter [1 to 4] of Perspectives for the energy transition – investment needs for a low-carbon energy system ©OECD/IEA and IRENA 2017.

Okanlawon SA, Olorumisola AO (2017). Development of passive evaporative cooling systems for tomatoes Part 1: construction material characterization. Agricultural Engineering International: CIGR Journal 19(1):178-186.

Parfitt J, Barthe M, Macnaughton S (2010). Food waste within food supply chains: quantification and potential for change to 2050. Philosophical Transactions of the Royal Society B-Biological Sciences 365:3065-3081.

Pathare PB, Opara UL, Vigneault C, Delele MA, Al-Said FA (2012). Design of packaging vents for cooling fresh horticultural produce: Review paper. Food Bioprocess Technology 5:2031-2045.
Paull RE (1999). Effect of temperature and relative humidity on fresh commodity quality. Postharvest Biology and Technology 15:263-277.
Paull RE, Duarte O (2011). Tropical fruits, CAB International.
Prusky D (2011). Reduction of the incidence of postharvest quality losses, and future prospects. Food Security 3(4):463-474.
Rayaguru K, Khan MK, Sahoo NR (2010). Water use optimisation in zero energy cool chambers for short-term storage of FV in coastal area. Food Science Technology 47(4):437-441.
Rennie T, Vigneault C, DeEll JR, Raghavan GSV (2003). Cooling and Storage. Handbook of Postharvest Technology: Cereals, fruits, vegetables, tea and spices. Ed. Chakraverty MA, Raghavan GSV, Ramaswamy HS. New York (NY), USA: Marcel Dekker Inc. pp. 505-538.
Said SAM, El-Shaarawi MAI, Siddiqui MU (2012). Alternative designs for a 24-h operating solar-powered absorption refrigeration technology. International Journal on Refrigeration. Available at: https://.doi.org/10.1016/j.ijrefrig.2012.06.008.
Samira A, Woldetsadik K, Workneh TS (2011). Postharvest quality and shelf life of some hot pepper varieties. Journal of Food Science Technology. Available at: https://doi.1007/s13197-011-0405-1.
Sandhya (2010). Modified atmosphere packaging of fresh produce: Current status and future needs. LWT-Food Science and Technology 43:381-392.
Saran S, Roy SK, Kitinoja L (2012). Appropriate postharvest technologies for small-scale horticultural farmers and marketers in sub-Saharan Africa and South Asia – Part 2. Field trial results and identification of research needs for selected crops. Proc. XXVIIIth IHC-IS on Postharvest Technology in the Global Market Eds.: M.I. Cantwell and D.P.F. Almeida. Acta Horticulture 934:41-52.
Sawicka B (2019). Postharvest losses of agricultural produce. Sustainable development. Available at: https://.doi:10.1007/978-3-319-69626-3_40-1
SAYB (2015). South African Year Book 2014/15, Agriculture. Department of Communication and Information System. Republic of South Africa.
Saxena A, Agarwal N, Srivastava G (2013). Design and Performance of solar air heater with long term heat storage. International Journal of Heat and Mass Transfer 60:8-16.
Schalkwyk HD, Groenewald JA, Fraser GC, Obi A, Tilburg A (2012). Unlocking markets to smallholders: Lessons from South Africa Mansholt publication series - l, 10. Available at: https://doi.org.3920/978-90-8686-168-2.
Senol R (2012). An analysis of solar energy and irrigation systems in Turkey. Energy Policy 47:478-486.
Shahzad MK, Chaudhary GQ, Ali M, Sheikh NA, Khalil MS, UrRashid T (2018). Experimental evaluation of a solid desiccant system integrated with cross flow Maisotsenko cycle evaporative cooler. Applied Thermal Engineering 128: 1476-1487. Available at: https://doi.org/10.1016/j.applthermaleng.2017.09.105.
Sibanda S (2019). Development of a solar powered indirect air-cooling combined with direct evaporative cooling system for storage of horticultural crops in hot and humid areas. PhD Thesis; University of KwaZulu Natal, Pietermaritzburg, South Africa.
Sibanda S, Workneh TS (2019). Effects of Indirect air-cooling combined with direct evaporative cooling on the quality of stored tomato fruit. CyTA Journal of Food 17(1):603-612. Available at: https://doi.org/10.1080/19476337.2019.1622595.
Sibanda S, Workneh, TS (2020). Performance of Indirect Air Cooling Combined with Direct Evaporative Cooling for Fresh Produce. Heliyon Journal. Available at: https://doi.org/10.1016/j.heliyon.2020.e03286
Sibomana MS, Workneh TS, Audain K (2016). A review of postharvest handling and losses in the fresh tomato supply chain: a focus on sub-Saharan Africa. Journal of Food Security 8:389-404. Available at: https://doi.org.1007/s12571-016-0562-1.
Sibomana MS, Ziena LW, Schmidt S (2017). Influence of transportation conditions and postharvest disinfection treatments on microbiological quality of fresh market tomatoes (cv. Nemo-netta) in a South African supply chain. Journal of Food Protection 80(2):345-354.
Singh V, Hedayetullah M, Zaman P, Meher J (2014). Postharvest technology of fruit and vegetables: An Overview. Journal of Post-Harvest Technology 2:124-135.
Singh D, Sharma RR (2018). Postharvest diseases of fruits and vegetables. In. Siddiqui (Ed) Postharvest disinfection of fruit and vegetables. Academic Press 1:1-52.
Sirisomboon P, Tanaka M, Kojima T (2011). Evaluation of tomato textural mechanical properties. Journal of Food Engineering 111(4):618-624.
Sitorus T, Ambarita H, Ariani F, Sitepu T (2018). Performance of the natural cooler to keep the freshness of vegetables and fruits in Medan City. IOP Conference Series: Materials Science and Engineering, 309, 012089. Available at: https://doi-10.1088/1757-899X/309/1/012089.
Sontake VC, Kalamkar VR (2016). Solar photovoltaic water pumping system - A comprehensive review. Renewable and Sustainable Energy Reviews 59:1038-1067.
Stathers T (2017). Quantifying postharvest losses in Sub-Saharan Africa with focus on cereals and pulses. [Internet]. Presentation at the Bellagio workshop on Postharvest management 12-14 September 2017. Available at: http://www.fao.org/fileadmin/user_upload/food-loss-reduction/Bellagio/T.Stathers_QuantifyingPHLinSSA.PDF. [Accessed 03 February 2020].
Sun W, Wang L (2004). Experimental investigation of performance of vacuum cooling for commercial large cooked meat joints. Journal of Food Engineering 61:(4) 527-532.
Sun W, Zheng L (2006). Vacuum cooling technology for the agri-food industry: Past, present and future. Food engineering 77:203-214.
Swain MJ, Evans JE, James SJ (2009). Energy consumption in the UK food chill chain–primary chilling. Food Manufacturing Efficiency 2(2):25-33.
Szabo S, Bódis K, Huld T, Moner-Girona M (2011). Energy solutions in rural Africa: mapping electrification costs of distributed solar and diesel generation versus grid extension. Environmental Research Letters 6(3):1-9.
Thompson JF, Mitchell FG, Rumsey TR, Kasmire RF, Crisoto CC (1998). Commercial cooling of fruits, vegetables and flowers. USA: DANR publication, UC Davis. Publication No. 21567:61-68.
Tigist M, Workneh TS, Woldetsadik K (2011). Effects of variety on quality of tomato stored under ambient temperature conditions. Food Science Technology. Available at: https://doi.org.1007/s13197-011-0378-0.
Tijskens E (2007). Impact damage of apples during transport and handling. Post-harvest Biology Technology 45(2):157-167.
Tiwari GN, Jain D (2001). Modelling and optimal design of evaporative cooling system in controlled environment greenhouse. Energy Conversion and Management 43:2235-2250.
Tolesa GN, Workneh TS (2017). Influence of storage environment, maturity stage and pre-storage disinfection treatments on tomato fruit quality during winter in KwaZulu-Natal, South Africa. Journal of Food Science and Technology 54(10):3230-3242. Available at: https://doi.10.1007/s13197-017-2766-6.
Tscharntke, T, Milder JC, Schroth G, Clough Y, DeClerck F, Waldron A, Rice R, Ghazoul J (2015). Conserving biodiversity through certification of tropical agroforestry crops at local and landscape scales. Journal of the Society for Conservation Biology 8(1):14-23. Available at: https://doi.org.1111/conl.12110.
Turk R, Celik E (1993). The effects of vacuum precooling on the half cooling period and quality characteristics of iceberg lettuce. Acta Horticulture 343: 321-324.
Tyagi VW, Panwar NL, Rahim NA, Kothari R (2012). Review on solar air heating system with and without thermal energy storage. Renewable and Sustainable Energy Reviews 16:2289-2303.
Ugonna CU, Jolaoso MA, Onwualu AP (2015). Tomato value chain in Nigeria: issues, challenges and strategies. Journal of Scientific and Reports 7(7): 501-515.
Van Zeebroeck M, Van linden V, Ramon H, De Baerdemaeker J, Nicolaï BM, Tijskens E (2007). Impact damage of apples during transport and handling. Post-harvest Biology Technology 45(2):157-167.
Vigneault C, Sargent SS, Bartz JA (2000). Postharvest decay risk associated with hydro-cooling tomatoes. Plant Disease 84(12):1314-1318.
Vigneault C, Thompson J, Wu S (2009). Designing container for

handling fresh horticultural produce. Postharvest Technologies for Horticultural Crops 2:25-47.
Wang LJ, Sun DW (2001). Rapid cooling of porous and moisture foods by using vacuum cooling technology. Trends in Food science and Technology 12:174-184.
Wills R, Glasson M, Graham D, Joyce D (2007). Postharvest: An Introduction to the physiology and handling of fruit, vegetables and ornamentals, (4th edition). New York, USA: University of New South Wales Press
Wilson LG, Boyette MD, Estes EA (1999). Postharvest handling and cooling of fresh fruits, vegetables and flowers for small farms. Horticulture information leaflets. USA: North Carolina Cooperative Extension Service 17:800-804.
Workneh TS (2007). Present status and future prospects of postharvest preservation technology of fresh fruit and vegetables in Ethiopia. Journal of the Ethiopian Society of Chemical Engineers 10(1):1-11.
Workneh TS (2010). Feasibility and economic evaluation of low-cost
evaporative cooling system in fruit and vegetables storage. African Journal of Food Agriculture, Nutrition and Development 10(8):2984-2997.
Workneh TS, Osthoff G (2010). A review on integrated agro-technology of vegetables. African Journal of Biotechnology 9(54):9307-9327.
Yahia EM (2011). Modified and controlled atmospheres for the storage, transportation, and packaging of horticultural commodities. Boca Raton, Florida: CRC press.
Zagory D, Kader AA (1988). Modified atmosphere packaging of fresh produce. Food Technology 70-77.
Zhang Z, Sun DW (2006). Effect of cooling methods on the cooling efficiencies and qualities of cooked broccoli and carrot slices. Journal of Food Engineering 77:320-326.
Zheng LY, Sun DW (2006). Innovative Applications of power ultrasound during food freezing processes - A Review. Trends in Food Science and Technology 17(1):16-23.
Zou Q, Opara LU, McKibbin R (2006). A CFD modeling system for airflow and heat transfer in ventilated packaging for fresh foods: II. Computational solution, software development, and model testing. Journal of Food Engineering 77(4):1048-1058.

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Risk assessment of the economic efficiency of rubber production: Case of smallholder rubber production in QuangBinh Province, Vietnam

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The paper reviews the economic risk of rubber production due to fluctuations of input factors, product price, natural disasters, epidemics, interest rates, and market demand. The data collected in this work were analyzed with accounting of production costs methods and accounting results and production efficiency method, benefit-cost analysis, sensitivity analysis and scenario analysis. QuangBinh, a province in Vietnam was used as the case study of this research; the area has great potential to develop smallholders producing rubber. QuangBinh rubber smallholders play an important role in developing local economy and creating jobs. However, this model has been facing risks, leading to a low economic efficiency. The study evaluates the overall economic efficiency of smallholder rubber production and business in Quang Binh Province in the context of risks: product price risk and interest rate risk. It also analyzes the relationship between selling price and interest rate with economic efficiency of smallholder rubber production.

Key words: Economic efficiency, economic efficiency in the context of risks, risks, risks of rubber, rubber, smallholder rubber production.

INTRODUCTION

In recent years, the world economy has been volatile coupled with the adverse effects of climate change that have caused many risks and reduced economic efficiency in agricultural business and rubber business. In particular, many scholars have carried out research on risks and economic efficiency in different areas. Research on risks in agricultural production has been done in three areas: The impact of natural factors (natural disasters, climate change ...), input factors (varieties, fertilizers, soil, farmers' qualifications, etc.) and the government impact factors on policies and legal framework. Several other studies address how to control risk in agricultural production with methods such as insuranceindex, risk classification and risk management tools. This study evaluates the factors that affect the efficiency of rubber production and business, factors leading to inefficient rubber production, economic efficiency analysis methods and policies to improve economic efficiency. Although there have been a lot of research on risks and economic efficiency in rubber

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production, there has not been any that aims to build a theoretical framework for assessing economic efficiency in relation to risks. Therefore, the paper aims to assess the economic efficiency of rubber production in the context of high risks, including factors responsible for risk like selling prices and interest rates, using QuangBinh Province, a locality in Vietnam as a case study.

MATERIALS AND METHODS

Based on the theoretical and practical research on risks and economic efficiency in rubber and smallholders' rubber production, the paper identifies the theoretical basis of smallholder rubber, rubber production risks and economic efficiency of rubber production. In particular, smallholder rubber is defined as a form of small-scale production; a situation in which farmers' households invest capital or lend money to farmers to invest in production and business development. In smallholder farms (with less than 4 haper household) plants are often scattered in them; the farms are scattered around where the farmers live (Dinh, 2000).A number of authors define risk as, "Risk is a measurable uncertainty" (Frank Knight); "Risk is uncertainty that may be related to the occurrence of unexpected changes" (Allan Willett); "Risk is a random sum that can be measured by probability" (Irving Preffer) or "Risk is a value and outcome that is currently unknown" . According to Arthur and Micheal, "Risk is the potential volatility in the results; occurs in almost every human activity, when there is a risk one cannot predict the exact outcome. The presence of risk causes uncertainty, risk can occur at any time, an action leading to unpredictability or loss" (Bui, 2005; Doan, 2013). In this study, the author determines that rubber production risks are events not intended by the producers such as natural disasters, epidemics, price fluctuations, changes in laws, farming techniques and measurable damage to rubber business results and efficiency. Regarding economic efficiency, according to Richard (1984), economic efficiency is the effective allocation of resources to bring surpluses to producers and consumers.

Economic efficiency is allocated based on the following conditions: (i) the value of one type of goods produced by an individual must be equal to the price of converting one commodity into another, (ii) the consumption value of the direct factors must be equal to the cost of converting the inputs into goods, (iii) the values are determined by the consumers of the inputs, and the output should be equal to the marginal products (Richard, 1984). This study identifies the economic efficiency of production and business of smallholder rubber plantation as a category; it reflects the comparative relationship between economic results and economic costs which households growing rubber spend to achieve in the rubber production and business cycle. Economic efficiency evaluation of rubber production is usually based on annual economic accounting indicators and long-term investment analysis criteria. However, to assess economic efficiency in the context of risk, it is necessary to analyze the fluctuations of the long-term indicators in terms of negative or positive correlation with risks. Therefore, the study uses sensitivity analysis method. Sensitivity analysis is an analysis of the relationship between insecure input quantities and output quantities. In other words, sensitivity analysis aimsto identify parameters that have a significant effect on the feasibility of a project and quantifying this level of influence. The analysis is to change the value of a project parameter and re-run the appraisal model to see how the NPV, IRR and evaluation criteria change. Sensitivity analysis in the risk assessment of rubber production and business is the determination of the variables thathave the most impact on the net benefits of rubber production and business and quantify their influence. This analysis includes testing the impact of the variation of selected costs and benefits on IRR and NPV of the rubber business production model.To accomplish the research objectives, the study uses the followings research methods:

Method of collecting data

Secondary data are the documents published through books, newspapers, summation reports and results of research works related to risks and economic efficiency of rubber production and business. Primary data are collected through questionnaire for smallholder rubber households. The sample size of the survey is determined by the following formula:

$$n = \frac{\mathrm{N}}{1 + \mathrm{N}(e)^2}$$

With the confidence of 95% and P = 0.5, the sample size with permissible error was ± 5%. The sample investigated was identified as n = 195; the number of households surveyed was 200 households. The data were collected according to the statistical method of stratification based on the life cycle of rubber trees. The surveyed households were selected randomly at each survey location.

Accounting of production costs

Determining the cost of rubber production was determined in 2 periods: the period of basic construction and the period of business. The study identified a basic construction period of 7 years from the year of planting rubber, including reclamation costs, new planting and interest expenses if any. Business period is from the 8th year; the cost includes labor cost, fertilizer cost, cost of production tools, depreciation cost of garden (all expenses for basic construction period allocated to years of economic periods) and financial expenses.

Accounting results and production efficiency

The actual yield of latex harvested for 1 ha of rubber of the surveyed household was calculated. The productivity data are usually obtain edvia interviews, household interviews and consideration of the statistical data on the productivity of the adjacent year (year) of the statistical office. The results and efficiency criteria are calculated as follows (Richard, 1984):

$GO = Q_i * P_i$

Where:
GO: Revenue collected per hectare of rubber tree area (VND 1,000)
Qi: Latex output of one hectare of rubber (kg)
Pi: Price of 1 kg of latex (VND 1,000)

Intermediate cost (IC)

This is the total amount of regular expenses spent to buy and rent inputs and services during the production of the total product.

Value-added (VA)

The value of products created during the manufacturing period. It is the difference between the value of production and the intermediate cost.

VA = GO – IC

Mixed income (MI)

This is the remaining added value after deducting expenses: depreciation of fixed assets, taxes and fees (if any).

MI = VA - depreciation of fixed assets - Tax - Bank interest (if any)

Profit:

Profit = MI - expense of family labor - expense of in kind of a household

Evaluation of production efficiency through indicators: GO / IC, MI / IC, LN / IC, VA / IC.

Benefit-cost analysis method: The research uses the benefit-cost analysis method in two ways: (1) Analyzing the annual cost benefit for the business period. The data used are primary data. Annual costs include expenses incurred during the year such as costs of supplies, production tools, labor, allocated depreciation of gardens and allocated financial expenses. Basic construction expenses are evenly distributed among the years of the business period; (2) cost benefit analysis for the entire production cycle, using NPV, BCR and IRR indicators. Benefits and costs arising in different years are realized at reasonable discount rates. The criteria are calculated by the following formula:

$$\text{NPV} = \sum_{t=0}^{n} B_t \frac{1}{(1+r)^t} - \sum_{t-o}^{n} C_t \frac{1}{(1+r)^t}$$

Where:
n: Number of life cycles of a rubber tree
t: Year of investment
Bt: Benefits of rubber trees in year t
Ct: Cost of rubber tree year t
r: Discount interest rate (% / year)
If NPV is > 0, the investment in rubber business is effective and profitable. Conversely, if NPV is <0, financially, this investment is ineffective and should not be implemented.

$$\text{IRR} = r_1 + (r_2 - r_1)\frac{NPV_1}{|NPV_1|+|NPV_2|}$$

Where
r_1: Lower discount rate at which NPV1> 0 is closest to 0
r_2: Higher discount rate at which NPV2 <0 is closest to 0.
NPV: Actual present value
The IRR to find (corresponding to NPV = 0) will lie between r_1 and r_2 IRR is the discount rate that makes NPV = 0; with this discount rate, rubber plantation is completely unprofitable because the income is just enough to offset the costs. Conversely, if IRR is greater than the interest rate, then production is efficient. The larger the IRR is, the higher the economic efficiency is.

Sensitivity analysis

Two variables were analyzed: price and lending interest rate.These are the two factors that have the biggest impact on the economic efficiency of rubber production. On that basis, the NPV value corresponding to the variable prices and interest rates that vary with a certain percentage was determined. Specifically, NPV was determined according to 2019 survey data, rubber prices and interest rates for the period 2014 - 2019. The analysis results are illustrated through graphs to assess the increase or decrease of NPV values when prices and interest rate change.

Scenario analysis method

Based on practical experience, this work proposes the situations for risk variables (input variables affecting production activities). This is to consider the change of a result variable and consider the change of economic efficiency criteria for smallholder rubber production when there are changes of many risk factors at the same time.

RESULTS

Assessing the impact of risk on the profit of rubber growers

The synthesis results in Table 1 show the risk of natural disasters causes maximum damage followed by cultivation, pests and diseases, product variety and price risks

Economic efficiency of rubber smallholdings in relation to product price risk

In Figure 1, at interest rates of 9%, the possibility for NPV of 01 hectares of rubber plantation under smallholder model to be greater than 0 is quite high and reaches 98%. Considering the direct relationship between price and NPV, price has a strong impact on change of NPV.With an estimated price change from VND 6,000/kg to 20,000/kg, NPV values change from VND 28.495 to 383.706 and the average value is VND181.416. Rubber latex prices are related directly to the NPV value obtained and the dependence of NPV on the price level is high.

Economic efficiency of smallholder rubber model in relation to loan rate risk

The NPV analytical results of smallholder rubber model in QuangBinhat different lending rates from 2013 - 2019 and of rubber latex price in 2019(10,000VND/kg) (Figure 2) shows that the interest rate has a direct influence on the value of NPV obtained. However, NPV value fluctuates due to interest rates, ranging from 1,168VND to 79,781VND. This range is lower than the impact of price, which is at VND10.000; NPV is greater than 0 in all cases of interest rate in stages from 2013 to 2019.NPV analysis, according to interest rate fluctuations and price in the period 2013- 2019, provides positive results. Most of the NPVs obtained from smallholder rubber model are greater than 0 in all cases. When looking at some specific NPV, in which farmers can decide the point at NPV is

Table 1. Economics efficiency of smallholder rubber business under risky conditions.

Type of risks	Frequency (persons)	Proportion (%)	Impact on profit (%)		
			Maximum	Minimum	Mean
Natural disaster, weather	200	100	100	4	26.83
Pest	200	100	30	1	8.28
Seedling	177	88.5	20	1	5.11
Technical cultivation	197	98.5	35	2	12.26
Seedling price	177	88.5	5	0.03	0.84
Pesticide price	200	100	7	0.09	1.5
Fertilizer price	200	100	7	0.1	1.6
Labor cost	200	100	10	0.2	2.2
Selling price	198	99	20	1	7.54
Changing in demand	0	0	-	-	-
Lacking of capital	200	100	15	0.5	6.54
Interest rate increase	200	100	17	0.5	6.34

Source: Survey results and calculations in 2019.

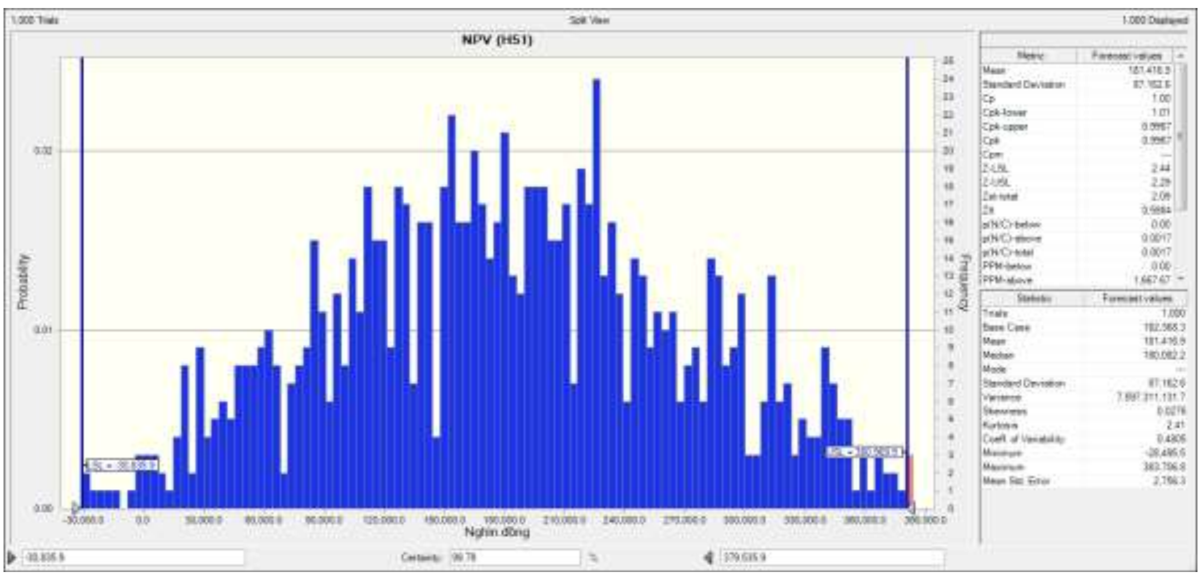

Figure 1. NPV distribution of smallholder rubber according to variation of rubber latex price from 2013 to 2019.

greater than 50%, it could be concluded that despite the risk, business will still be profitable if the level of prices and interest rates is fluctuating around 2019.

Economic efficiency in relation to product price and loan rate risks

NPV analysis results show the variability of latex price and interest rates from 2013-2019 with latex prices from VND 7,000/kg or more. The NPV in this case is positive; even with the highest interest rate at 17.5% in 2014, the NPV is still higher than 0 in all cases of more VND 10,000 /kg for rubber latex. However, current prices are maintained at VND 10,000/kg, so the NPV is positive (Table 2). There are many factors affecting the business of rubber production such as price, interest rate, etc. However, at 7,000VND/kg prices or higher and interest rates at 17.5% or less, the NPV in all cases would be positive. These show that the investment in small holder rubber production in QuangBinh Province has high economic efficiency.

Analyzing the relationship between selling price and lending interest rate with economic efficiency of smallholder rubber production

The relationship between the product price and interest rate changes compares well with prices and interest rates

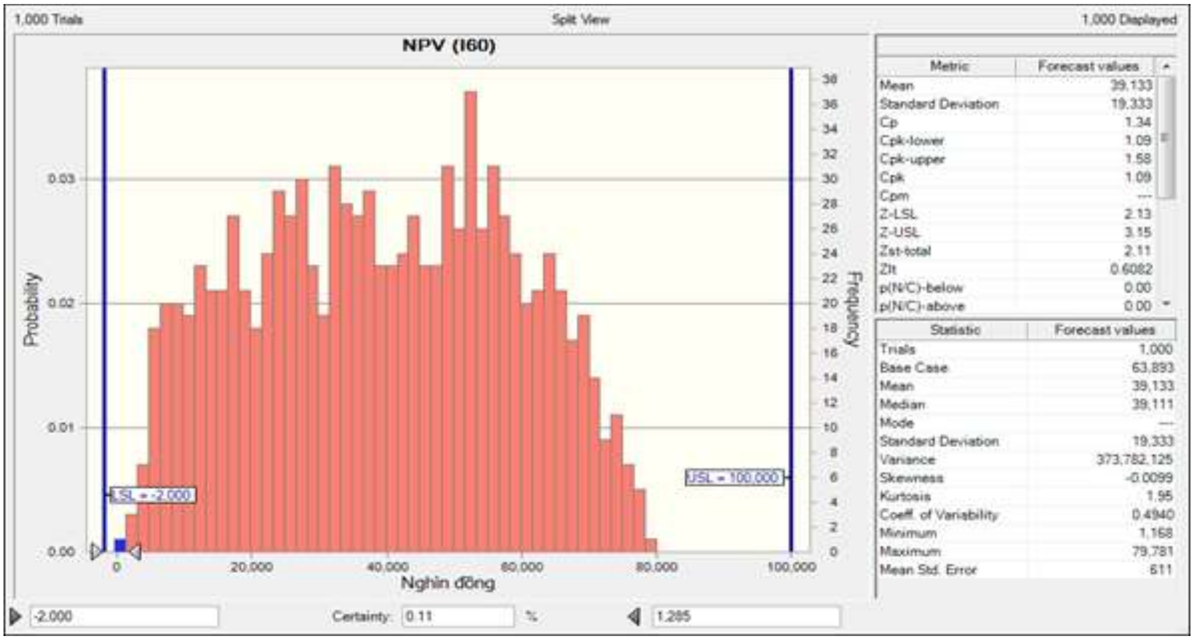

Figure 2. Variation of NPV value according to interest rate in stages from2013 to 2019.

Table 2. Sensitivity of NPV as prices and interest rates vary from 2013 to 2019.

Price (Thousand VND) / Rate (%)	6.2	6.9	13.0	20.3	13.5	10.8	10
17.0	-35,483	-28,543	35,833	112,528	41,086	12,720	4,315
17.5	-35,609	-28,255	31,940	104,321	36,897	10,127	2,194
11.5	-34,498	-19,958	106,747	258,377	117,132	61,050	44,433
12.5	-35,208	-22,447	88,754	221,831	97,869	48,648	34,065
15.0	-35,845	-26,515	54,794	152,098	61,459	25,470	14,806
10.0	-32,697	-14,904	140,148	325,702	152,857	84,227	63,893
9	-30,836	-10,392	167,765	380,970	182,368	103,512	80,147

Source: Survey results and calculations in 2019.

according to survey data of VND 10,000 and 9%. The results in Table 3 show that NPV, IRR and BCR are quite sensitive to changes of inputs. However, the ability to leave NPV <0 in all situations is quite low. With the increase or decrease of the price, as well as the change of the discount rate, NPV in all cases is NPV> 0. In other words, when interest rates and prices change around the scenario level in 2019, they do not suffer from losses. When prices fall from 10-20% this directly affects sales; this scenario leads to a significant decline of NPV, IRR, BCR.

The analytical results show that, even when the interest rate is much reduced (20%), smallholder rubber production is still profitable. Meanwhile, with the increase in price, there is increase in sales; the NPV, IRR and BCR also increase. For example, with a price increase of 20%, the NPV increases by more than 58,000 and the BCR index increases by 0.27 times compared to the scenario in 2019. Similarly, the decrease in interest rates also directly affects the NPV and BCR. However, for IRR, the change is not too significant. Thus, when compared to the 2019 scenario, although the indicators change with the increase and decrease of price factors as well as discount rates, NPV results are always > 0.

DISCUSSION

A lot of studies have discussed risks and economic efficiency in rubber business. Claire (2010) says that the

Table 3. Summary of CBA criteria for QuangBinh smallholder rubber plantation by scenario.

Scenario	NPV (1.000vnd/ha)	IRR (%)	BCR (times)
Price at VND 10,000 and interest rate of 9% (2019)	80.147	18	1.36
1. Price changes, interest rate 9%			
Downto 10%	50.941	15.3	1.23
Down to 15%	36.338	13.7	1.17
Down to 20%	21.735	12.02	1.10
Up to 10%	109.353	20.4	1.50
Up to 15%	123.956	21.46	1.56
Up to 20%	138.559	22.19	1.63
2. Interest rate changes, fixed price VND 10,000			
Up to 10%	65.387	18.00	1.33
Up to 15%	58.871	18.07	1.31
Up to 20%	52.864	18.00	1.29
Down to 10%	97.595	18.00	1.40

Source: Survey results and author calculations in 2019.

instability in agricultural production evolves in an increasingly complex direction. These instabilities may result from weather, price fluctuations, seasonal output, supply-demand correlation, energy price fluctuations. Sarba (2011) in studying the effectiveness of the production of natural rubber in Indonesia, identified that prices have a direct and large impact on farmers' income; Somboonsuke et al. (2002) commented that low rubber prices were a major obstacle for rubber growers. Jagath et al. (2010)in Sri Lanka focused on analyzing factors affecting inefficient production. The authors found that the main reason for reducing the economic efficiency of smallholder rubber production is the fluctuation of prices, which has a direct influence on the income of rubber growers. The study pointed to the ineffective use of inputs and concluded that there was no point in increasing inputs but use efficiency has to be improved to increase the profitability of the household. Sarba (2011) studying the factors affecting the growth of production capacity in the rubber industry, concluded that there is need for government's reformed policies on licensing trade liberalization. It does not help industry and agriculture to expand its capacity; most of them use their own production capacity and have not yet used up this capacity. Approaching other research methods, Ririnet al. (1999)assessed the importance of rubber trees in the Indonesian economy, including the price change affecting the profitability of growers. The authors also use a bio-economic model to assess production efficiency between the standard model and the second model after introducing risk factors.

Thus, in the study of theory and practice of risks and economic efficiency in rubber production and trading, the works mainly discussed each in different aspects. There are works that only study risk factors or risk control in agricultural production and risk analysis methods. Researching on the economic efficiency of rubber production, researchers refer to the history of rubber and rubber plantation formation and development, the role of rubber plantation and the factors affecting production and business efficiency of rubber; factors leading to inefficient rubber production, economic efficiency analysis methods and policies to improve economic efficiency. There has not been any study to assess the economic efficiency of rubber production and business in relation to risks. Therefore, thisstudy assesses the economic efficiency of rubber production in the context of risks.Smallholder rubber production in Quang Binh province, Vietnam ensures novelty and creativity. The work uses the common theory to form the theoretical basis of smallholder rubber model, risks in rubber production and assess the economic efficiency of rubber production, thereby forming methodsto study and evaluate the economic efficiency of rubber production and business in the context of risks. It evaluates the model of smallholder rubber in Quang Binh province, Vietnam. The research results have supplemented and enriched the theory of risk and assessed economic efficiency in rubber production and business; it is an important source of reference for organizations and individuals to study and implement issues related to rubber and rubber production and business. At the same time, they are important practical bases to help smallholder rubber households and local authorities at all levels in Quang Binh Province have strategies and solutions to minimize risks and improve economic efficiency.

Conclusion

Small-scale rubber production has many fluctuations, long production cycles, and different annual productivity;

so if based on the results of assessing economic efficiency at a time, it will lead to risks for producers. Therefore, the evaluation of economic efficiency of smallholder rubber production in the context of risk is the assessment of economic efficiency under uncertain future conditions; so the results of the research reflect the most comprehensive and accurate expression. The article has studied systematizing the theory and practice of risk research, assessing economic efficiency in rubber plantation production. Then, the concepts of smallholder rubber, risks of rubber production, economic efficiency of rubber production and sensitivity analysis method have been used as a basis for theoretical research. The results of the study showed that smallholders in rubber without risk conditions will be highly effective, in the context of reduced profitability and reduced efficiency with the level of each type of risk.

Risk from strong storms has the greatest impact that can reduce profits by 100% and, on average, reduce profits by 26.8%. Other risks have lower impacts; but there are some with more significant impacts such as the risk of falling product prices, rising interest rates or risks due to unsecured farming techniques. Studying the context of the current risks in QuangBinh province, investing in smallholder rubber is effective; the author analyzes the fluctuation of NPV value by latex price and interest rate in the period of 2013-2019. It shows that, with prices ranging from 7,000 VND/1kg of fresh latex or more and interest rates ranging from 17.5% or less, NPV in all cases are positive. At the same time, analyzing the scenario of selling price and lending interest rate shows that the indicators NPV, BCR and IRR all change according to the increase and decrease of price factors as well as discount rates but NPV always achieve results> 0 The results of this analysis show that in the current risks, investment in smallholder rubber production and business in QuangBinh province is still effective.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Bui TG (2005). Risk management in agricultural production and business establishments. University of Agriculture - Hanoi, Vietnam.

Dinh XT (2000). Researching models of monetary policy in Vietnam, Final report, Vietnam Rubber Research Institute.

Doan TH (2013). Risk and crisis management, Labor - Social Publishing House, Hanoi, Vietnam.

Claire SC (2010).Risk Management in Agriculture, Science and Education. An Open Access and Academic Publisher. https://www.dbresearch.com/PROD/RPS_EN-PROD/PROD0000000000466866/Risk_management_in_agriculture%3A_Towards_market_sol.PDF

Jagath E, Wasana W,Bogahawatte C(2010).Profit efficiency of smallholder rubber farmers in Kegalle, Kalutara and Ratnapura districts. Journal of the Rubber Research Institute of Sri Lanka 90:64-78.

Richard LK (1984). Methods for Evaluating Economic Efficiency in Agricultural Marketing.Journal of Agricultural and Applied Economics 16:1.

Ririn P, Oscar C, Phil S (1999). Management strategies for indonesia small – holder production in south Sumatra: a bioeconomic analysis. Working Papers 12936, University of New England, School of EconomicsNo.99 – 14.https://ageconsearch.umn.edu/record/12936/

Sarba PR (2011).Economic Efficiency in Indian Rubber Industry. Contemporary Economics 5:82-89.

Somboonsuke B, Shivakoti GP, Demaine H (2002). Rubber-based farming systems in Thailand: Problems, potential, solutions and constrains. Journal of Rural Development 21(1):85-113.

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Nitrogen value of stockpiled cattle manure for crop production

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Cattle manure is a common source of fertilizer throughout Sub-Saharan Africa given problems with supply and pricing of inorganic fertilizers. The optimum rate of manure to use is often unknown and further compounded by variable N contents arising from long periods of stockpiling. This study investigated the rates of cattle manure required to optimize plant growth at different N contents arising from different storage times. A field experiment was established using cattle manure stored in the open for 4, 12 and 13 months (N content of 1.31, 1.18and 0.32%, respectively). Plant shoot dry weight and N uptake of canola (*Brassica napus* L) was compared to equivalent rates of inorganic N (urea) to 200 kg N ha^{-1}at two sampling points over two growing seasons. Linear models of the form y = a + bx were fitted to the data where y is yield (dry matter or N uptake) to enable the N equivalent value (NEV) of cattle manure treatments to be compared to inorganic fertilizer. The NEV of cattle manure stored for 4 months averaged 30% for shoot dry weight and 24% for N uptake and decreased with storage. Impractical volumes of cattle manure are required for plant production in aged stockpiles, thus necessitating better options for N management.

Key words: Sub-Saharan Africa, cattle manure, nitrogen fertilizer, N equivalent value, stockpiled manure.

INTRODUCTION

Nitrogen deficiency is among the top five factors limiting soil fertility in Sub-Saharan Africa (SSA) (Stewart et al., 2020). Studies by Muhereza et al. (2014) identified N as the limiting soil macro-nutrient for many farmers in peri-urban Uganda. The application of inorganic fertilizer by smallholder farmers in SSA to improve soil nutrient levels is low compared to world standards (Sheahan and Barret, 2014; Kaizzi et al., 2017). Numerous economic factors such as price policies and credit, distribution costs, the privatization of supply, and infrastructural development contribute to the low availability and use of inorganic fertiliser (Mwangi, 1996; Croppenstedt et al., 2003). Instead, cattle manure is commonly used by smallholder farmers to improve crop growth (Ndambi et al., 2019). Itis usual practice for cattle manure to be stored in piles for periods of between 2 to 6 months to

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Table 1. Percentage Nitrogen (N) content and N equivalent values (NEV) for a range of cattle manures and crop responses compared to inorganic N in the first year of application.

Manure type	N Content %	Crop	NEV%	Reference
Solid FYM	0.8	Silage	4-32	Chambers et al. (1999)
Solid manure	1.6	Corn	10-50	Gale et al. (2006)
Solid manure	0.7	Silage	35	Ferguson et al. (2005)
Solid manure	0.5	Barley and potato	46-59	Mallory et al. (2010)

bulk up adequate manure for the crop prior to application. The manure is sourced from farmers that practice animal confinement and manure storage (Muhereza, 2011; Katuromunda et al., 2012).

There are limited studies available to determine the volume of cattle manure required to supply adequate nutrients for crop growth, particularly where stockpiled over long periods of time. The nutrient content of cattle manure varies and is affected by various factors such as the type of livestock (Griffin et al., 2002; Esfahani et al., 2016), age, diet, the inclusion of bedding material, the duration and conditions of storage and the kind of treatment prior to soil application (Reijs et al., 2007;Snijders et al., 2013; Leip et al., 2019). The N content in cattle manure in SSA is low in N compared to elsewhere and is attributed to poor handling, collection and storage of manure, insufficient fodder and poor livestock diet. For example, values of 0.56% N have been measured in fresh manure in Uganda, with declines to 0.44% N in uncovered manure and 0.42% in aged manure (Muhereza, 2011). The N Equivalent Value (NEV) calculates the equivalent amount of N in manure required to give similar plant yield at comparable rates of inorganic N. Table1 shows that the NEV for cattle manures (N content from 0.5 to 2.1%) varies considerably from 0 to 59% in the first year of application compared to inorganic fertilizer N. The type of manure, for example solid manure, composted manure or fresh manure is responsible for much of this variation (Table 1).

Therefore, to better improve soil fertility and optimize food production where cattle manure is used as a source of fertilizer in SSA, research is required to determine effective rates of manure required following stockpiling for different lengths of time. This research aims to determine the effect of stockpiling of cattle manure on its value as a source of N. There search outcomes will better guide manure application rates and storage options and is relevant to developing countries with similar farming systems with limited access to fertilizer N.

MATERIALS AND METHODS

Experimental design

A field experiment was established over two growing seasons (2009 and 2010) using three sources of stock piled cattle manure, comprising manure stored in the open for 4, 12 and 13 months; designated as M_4, M_{12} and M_{13}, respectively. The manure was sourced from a feedlot where cattle had been fed a maintenance diet of pelletised hay and grain. The stored manure was of low quality to reflect manure commonly available to farmers in SSA. The concentration of total N in the stockpiled manure decreased with storage time, ranging from 1.31 to 0.32% (Table 2).

The 2009 experiment comprised 12 treatments; six (6) rates of stockpiled M_{13} cattle manure (0.32% total N) and 6 rates of inorganic N fertilizer. The 2010 experiment comprised 18 treatments; 6 rates of stockpiled M_4 cattle manure (1.31% total N), six (6) rates of stockpiled M_{12} cattle manure (1.18% total N) and 6 rates of inorganic N fertilizer (Table 2). The rates of inorganic N applied as urea (46% total N) up to 214 kg N ha^{-1}were selected to apply similar amounts of N to manure treatments (Table 3). All treatments included three replications to give a total of 36 plots in 2009 and 54 plots in 2010.Plots measuring 2m × 2m and were arranged in a systematic gradient design to prevent edge effects that may result from a high rate plot adjacent to a low rate plot as per Rigby (2008). The elimination of wide guard rows facilitated an increased number of fertilizer rates, increased the ratio of harvested to non-harvested area and reduced the potential threats from experimental error.

Location and soil type

The field experiments were established at Ucarty, located 120km east of Perth, Western Australia (-31.19159°S, 116.57083°E) on a dark yellowish-brown sand (Table 4).The region experiences a dry temperate climate with cool, wet winters and hot, dry summers, and an average rainfall of 302 mm over the growing season (April-October). The site was chosen to be low in N to enable a nutrient response to be determined.

Application of manure and basal nutrients

To ensure that all plant nutrient requirements were adequate other than N, basal nutrients were broadcast by hand to the site prior to planting and then incorporated by disc plough working across plots; 0.04 kg P ha^{-1} (superphosphate 9 kg ha^{-1}), 0.24 kg K ha^{-1} (muriate of potash 588 kg ha^{-1}), 0.003kg Cu kg ha^{-1} (copper sulphate 6.7 kg ha^{-1}), 0.005 kg Zn ha^{-1} (zinc sulphate 13.3 kg ha^{-1}), 0.01 kg $CaSO.2H_2O$ ha^{-1} (gypsum 12.5 kg ha^{-1}) and 0.05 kg Mnha^{-1} (manganese sulphate 120 kg ha^{-1}). Cattle manure was weighed into buckets using an electronic weighing scale, broadcast evenly by hand onto respective plots, and then incorporated into the soil during seeding by discs. Inorganic N was broadcasted by hand onto the respective plots and split over two applications at seeding and at 8 weeks after planting, a common practice in the region. The first application was incorporated at seeding by discs and the second

Table 2. Total percentage of nitrogen (N) in the three stockpiled cattle manures used in the study.

Analysis	Type of cattle manure		
Dry matter content	**M_4**	**M_{12}**	**M_{13}**
Total N (%)	1.31	1.18	0.32

Western Australian ChemCentre report # 09A0508:5. National Association of Testing Authorities - NATA accredited. Analysis measured on sub-samples at a depth of 45 cm from the surface of the stockpile. Cattle manure oven dried at 105°C in a forced air oven and reported as 90% dry basis, mg kg^{-1}. Total N by combustion. Nitrate-N and ammonium-N measured by SFA and less than 0.04 mg kg^{-1} in all samples. Other constituents in M_{13} as follows: pH 7 (0.01M $CaCl_2$; 1:5), organic carbon 6.7%W/B, 0.11 mg total P kg^{-1}.

Table 3. Target rates of N (kg ha^{-1}) and equivalent quantities of cattle manures (kg $plot^{-1}$) at three stockpile months applied in the study.

Target rate N (kg ha^{-1})	M_4 (kg $plot^{-1}$)	M_{12} (kg $plot^{-1}$)	M_{13} (kg $plot^{-1}$)
0	0	0	0
40	3.0	3.4	12.5
86	6.1	6.8	26.8
128	9.2	10.2	40.0
170	12.2	13.6	53.2
214	15.3	16.9	66.8

Cattle manure rates calculated at a DM of 90%.

Table 4. Selected soil characteristics of the <2 mm fraction of the top soil (0-10 cm) at the study site.

Parameter	Value
pH (0.01 M $CaCl_2$; 1:5)	5.0
EC (1:5) (mS/m)	6.0
Sand (%)	95.5
Silt (%)	2.0
Clay (%)	2.5
Organic C (%W/B)	0.74
Total N (%)	0.065
Total P (mg/kg)	74.0
P ($HCO_{3\ Colwell}$) (mg/kg) l	7.0
K ($HCO_{3\ Colwell}$) (mg/kg)	45.0

Western Australian ChemCentre WA Report 08A7/1-5 (National Association of Testing Authorities - NATA accredited) (n = 4). Values averaged from four random 5 cm diameter soil cores bulked and then dried at 40^0C for 72 h in a forced air oven.

application was top dressed only.

Seeding and site management

Canola (*Brassica napus* L) is efficient at capturing high amounts of N during vegetative growth (Kazemeini et al., 2010). It was suitable for the dryland temperate climate of the study site and is from a similar family to cabbage (*Brassica oleracea*), a staple food crop in Uganda. Canola has similar early plant growth and N uptake to canola (Muhereza, 2011).Canola was sown at the start of each growing season at a rate of 9 kg $ha^{-1}$by top dressing by hand and incorporating to a maximum depth of 4 cm by a disc seeder. Weeds were controlled by spraying with glyphosate (2 L ha^{-1}) prior to planting and Amor (150 mL ha^{-1}) for grass control five weeks after planting and by hand weeding on sampling days. An electric fence was erected to prevent cattle and field rabbits from damaging the crop. Weather data (maximum, minimum and average) was recorded by an automated weather station.

Plant growth measurements and nitrogen uptake

Whole plant (above ground) samples of canola were collected from 0.25 m^2 quadrants of each plot at 63 days after sowing (DAS)and 93DASin 2009 and 70 DAS and 100 DAS in 2010. Plant tissue was dried to a constant mass at 70°C for 48 h in a forced-air oven to determine dry matter (DM). The average plant density per plot was determined at the first sampling date. The dried plant samples (g m^{-2}) were then used to determine concentration of total Nin shoots by combustion in a Leco FP-428 analyser. Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-AES) was used to determine concentrations of other plant nutrients; P, K, Ca, Mg, Na, S, Fe, Mn, Zn, and Cu(CCWA. 2000). Nitrogen uptake by canola was determined by multiplying the shoot DM yields with the N concentration.

Data analysis

Statistical analysis of data was carried out using standard analysis of variance (ANOVA) (Gen Stat Procedure Library Release PL20.1) (Payne et al., 2011). The significance of the treatment effect was determined using the F-test, while the least significant differences (LSD) between the means of the six treatments were estimated at the 5% probability level to determine if cattle manure had a significant effect on crop growth and yield compared with inorganic fertilizer.

Dry matter yields and N uptake in cattle manure were analysed by linear regression analysis and compared to respective yields from mineral N calibration plots using models of the form $y = a + bx$ where y is the yield (kg ha^{-1} DM or N uptake). The quadratic equations of the form ($y = a + bx + cx^2$) was tested for the data but the crop response did not approach a curvilinear at the top rates of application and the quadratic term (x^2) in the crop response model was not significant at $P<0.05$. Hence linear regression was considered the most appropriate to use in this study, particularly as yield response to manure N is considered linear between applications of 0-300 kg N ha^{-1} (Whitehead, 1995).Where the linear regression was significant according to ANOVA analysis, NEV were obtained by comparing the slopes of the regression equations for N from cattle manure to the slope obtained for inorganic N for both DM yield and N uptake.

RESULTS

Weather

The growing season rainfall (May to October) at the study site received 246 mm in 2009 and 237 mm in 2010. Rainfall in both years of the experiment was relatively low compared to the long-term average of 302 mm, with low out of season rainfall experienced between the two years.

Canola dry matter yield

The DM yield of canola shoots increased with increasing rates of inorganic-N at all sampling dates for both seasons (Figure 1). Inorganic-N treatment at 200 kg N ha^{-1} reached a maximum yield of 2,582 kg DM ha^{-1} during the first season (2009) at 93 DAS and 1,400 kg DM ha^{-1} during the second season (2010) at 100 DAS. At all N rates and sampling dates, the application of inorganic-N resulted in higher yields compared to all sources of cattle manure-N at equivalent application of N (P<0.001). There were large increases in DM yield for inorganic-N between the rates of 40 kg N and 80 kg N ha^{-1}.

In cattle manure stockpiled for 13 months (M_{13}),the maximum DM yield of canola at 63 DAS was 190 kg ha^{-1}at the highest N application rate (214 kg N ha^{-1}) and increased to 631 kg ha^{-1}at 93 DAS (Figure 1). Shoot DM yields averaged 4 times less than equivalent inorganic-N over the growing season. In cattle manure stockpiled for 12 months (M_{12}), the maximum DM yield of canola at 70 DAS was80 kgha^{-1}at the highest N application rate and increased to 537 kgha^{-1} at 100 DAS (Figure 2). Shoot DM yields averaged 3 times less than equivalent inorganic N over the growing season. In cattle manure stockpiled for 4 months (M_4), the maximum DM yield of canola at 70 DAS was 110 kg ha^{-1} at highest N application rate and increased to 260 kg ha^{-1}at 100 DAS (Figure 2). The DM yield of canola in equivalent rates of inorganic-N was600 kg ha^{-1} at 70 DAS and increased to 1,436 kg ha^{-1} at 100 DAS. Compared to equivalent rates of inorganic-N, shoot DM yields were reduced from 2.3to 2.4 times less over the growing season.

Shoot nitrogen concentration and uptake

The concentration of N in the DM of canola shoots was positive to increasing rates of N (data not given). Over both growing seasons (2009 and 2010), shoot N concentrations declined overtime with overall means as follows; 3.2 to 2.8% from 63 to 93 DAS and 3.4 to 2.6% from 70 to 100 DAS, respectively. Mean total N uptake by canola shoots increased with increasing rates of either inorganic N or cattle manure N (M_4,M_{12} and M_{13}) at all sampling dates (Tables 4 and 5).The highest values of N uptake (kg ha-^{1}) were in the inorganic-N fertilizer treatments and ranged between 1.2 and 14.0 kg N ha^{-1}at the first sampling and between 5.0 and 54.5 kg ha-^{1}at the second sampling time as rates of N increased from 0 to 120 kg ha^{-1}. As the storage time of manure increased, the total N uptake by canola decreased, with manure stored for 13 months having the lowest N uptake compared to all other treatments at both sampling dates. There were significant differences (P<0.001) in N uptake between inorganic-N and cattle manure-N at comparable total N loadings at both sampling dates.

Stockpiled cattle manure-N compared with inorganic-N during the growing season

Nitrogen equivalent values (NEV) of stockpiled cattle

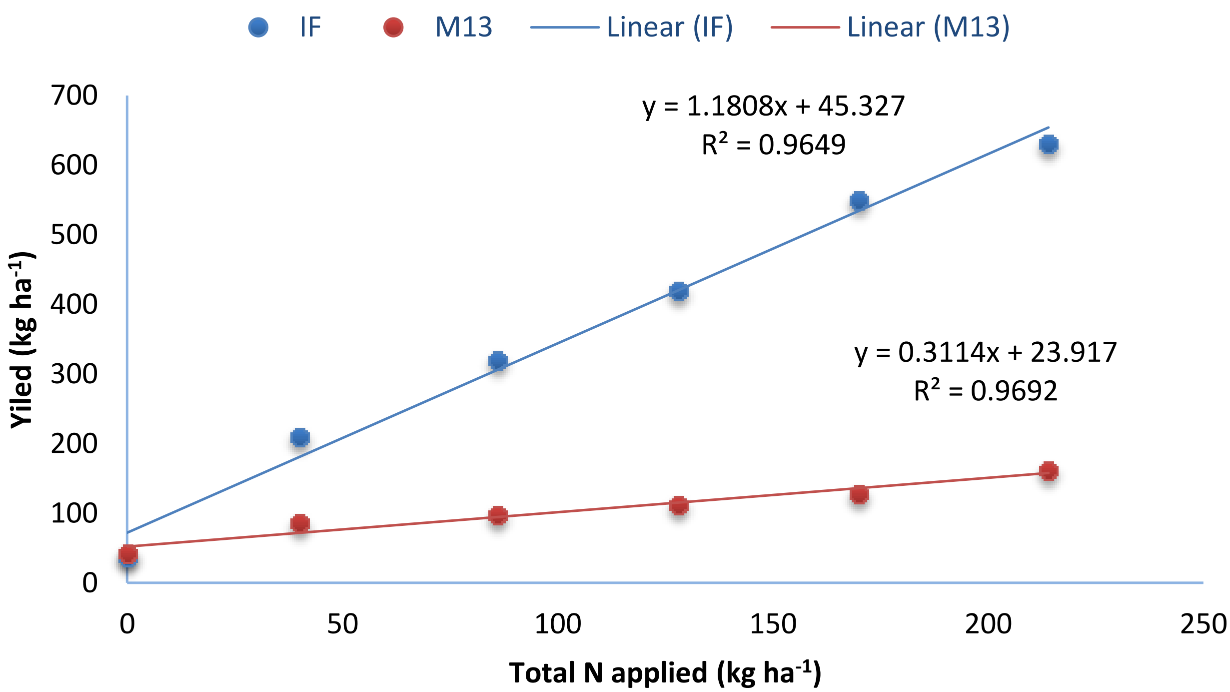

Figure 1. Mean vegetative dry matter yield (kg ha^{-1}) in relation to rate of total N application in inorganic-N (urea), cattle manure-N stored for 13 months (M_{13}) at 63 DAS with regression equation and standard error bars given. Values are the means of three replicates. Line plotted using Microsoft Excel. Vertical bars denote LSD (P = 0.05) for data from all treatment means.

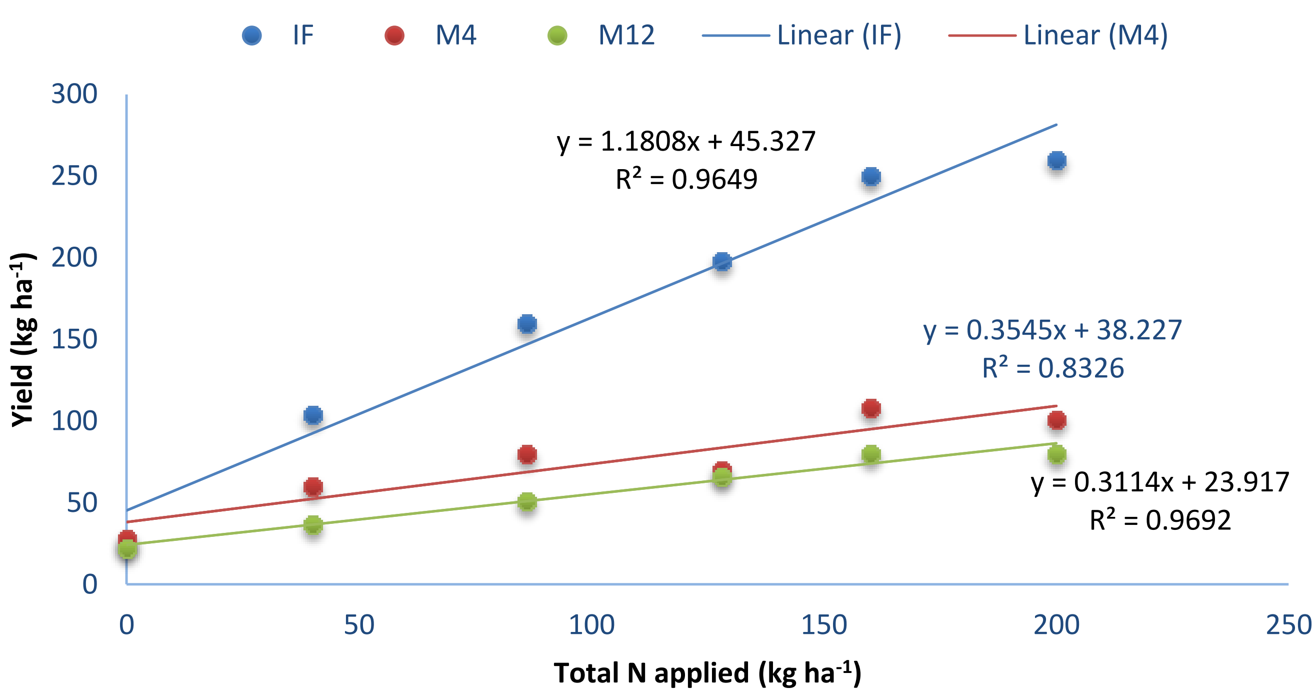

Figure 2. Mean vegetative dry matter yield (kg ha^{-1}) of canola in relation to rate of total N application in inorganic-N (urea), cattle manure-N stored for 4 months (M_4) and 12 months (M_{12}) at 70 DAS in 2010 with regression equation and standard error bars given. Values are the means of three replicates. Line plotted using Microsoft Excel. Vertical bars denote LSD (P = 0.05) for data from all treatment means.

Table 5. Mean total N uptake (kg ha-1) in canola shoots in relation to equivalent levels of N applied in inorganic fertilizer (IF) and stockpiled cattle manure at the first sampling period (between 63 and 70 DAS).

Source	N Rate (kg ha-1)					
	0	40	80	120	160	200
M_4	1.1	1.8	2.4	3.0	3.1	3.8
M_{12}	1.2	1.7	1.8	2.3	2.7	2.7
M_{13}	0.9	1.5	1.5	1.8	2.1	2.2
IF	1.2	3.4	6.6	8.7	11.1	14.0

$M_{4:}$ manure stored for four months (63 DAS); $M_{12:}$ manure stored for twelve months (70 DAS); $M_{13:}$ manure stored for thirteen months (70 DAS); $_{IF:}$ inorganic N fertilizer.

Table 6. Mean Total N uptake (kg ha^{-1}) in canola shoots in relation to equivalent levels of N applied in inorganic fertilizer (IF) and stockpiled cattle manure at the second sampling period (between 93 to 100 DAS).

Source	N Rate (kg ha-1)					
	0	40	80	120	160	200
M_4	5.0	8.1	8.7	11.3	16.2	18.7
M_{12}	5.0	8.2	10.3	9.9	13.2	15.9
M_{13}	4.0	6.5	8.0	9.2	11.4	13.0
IF	5.0	15.5	24.0	31.9	40.3	54.5

$M_{4:}$ manure stored for four months (93 DAS); $M_{12:}$ manure stored for twelve months (100 DAS); $M_{13:}$ manure stored for thirteen months (100 DAS); $_{IF:}$ inorganic N fertilizer.

Table 7. Linear regression coefficients and r^2 values for DM yield and N uptake in canola in stockpiled cattle manure-N (M_4, M_{12} and M_{13})and inorganic-N fertilizer (IF) at the first sampling times of 63 DAS (2009) or 70 DAS (2010).

Source	Dry weight				N Uptake			
	63 DAS		70 DAS		63 DAS		70 DAS	
	Slope	r^2	Slope	r^2	Slope	r^2	Slope	r^2
M_4	-	-	0.307	0.64	-	-	0.013	0.69
M_{12}	-	-	0.216	0.49	-	-	0.072	0.45
M_{13}	0.497	0.91	-	-	0.017	0.52	-	-
IF	3.036	0.98	1.117	0.74	0.127	0.85	0.064	0.81

$M_{4:}$ manure stored for four months; $M_{12:}$ manure stored for twelve months; $M_{13:}$ manure stored for thirteen months, $_{IF:}$ inorganic N fertilizer.

Table 8. Linear regression coefficients and r^2values for DM yield and N uptake in canola in stockpiled cattle manure-N (M_4, M_{12} and M_{13}) and inorganic-N fertilizer (IF) at the second sampling time of 93 DAS (2009) or 100 DAS (2010).

Source	Dry weight				N Uptake			
	93 DAS 2009		100 DAS 2010		93 DAS 2009		100 DAS 2010	
	Slope	r^2	Slope	r^2	Slope	r^2	Slope	r^2
M_4	-	-	1.99	0.63	-	-	0.07	0.68
M_{12}	-	-	1.54	0.71	-	-	0.05	0.64
M_{13}	2.29	0.86	-	-	0.06	0.69	-	-
IF	10.41	0.94	6.03	0.89	0.35	0.86	0.24	0.88

$M_{4:}$ manure stored for four months; $M_{12:}$ manure stored for twelve months; $M_{13:}$ manure stored for thirteen months, $_{IF:}$ inorganic N fertilizer.

manure compared with inorganic-N fertilizer for each measure of crop response (DM yield and N uptake) demonstrated a significant relationship between rates of N application ($P<0.05$) (Tables 7 and 8). The fitted modelaccounted for 64 to 98% of the variance of the experimentally derived results for DM at the first sampling

Table 9. Nitrogen equivalent values (NEV) of stockpiled cattle manure for DM yield (kg ha^{-1}) and N uptake (kg ha^{-1}) of canola on comparison of linear regression coefficients with inorganic-N fertilizer at two sampling dates for the two years.

Source	Total N (%)	DAS	Year	Dry matter (kg ha^{-1})	N uptake (kg ha^{-1})
M_4	1.31	70	2010	0.27	0.20
M_{12}	1.18	70	2010	0.19	0.11
M_{13}	0.32	63	2009	0.16	0.13
M_4	1.31	100	2010	0.33	0.28
M_{12}	1.18	100	2010	0.26	0.21
M_{13}	0.32	93	2009	0.22	0.18

DAS: Days after sowing; M_4: manure stored for 4 months at 1.31% total N; M_{12}: Manure stored for twelve months at 1.18% total N and M_{13}: Manure stored for thirteen months at 0.32% total N.

date. The slopes for DM of the fitted linear regressions were0.31, 0.22 and 0.50 for the three manure sources, M_4, M_{12} and M_{13}, respectively at the first sampling date (63 to 70 DAS) (Table 7). By the second sampling date (93 to 100 DAS), modelled values accounted for 63 to 94% with slopes of 1.99, 1.54, and 0.86 for the three manure sources, M_4, M_{12} and M_{13}, respectively.

The NEV for all sources of manure compared to inorganic N fertilizer was calculated for both DM and N uptake (Table 9). The NEV for DM at the first sampling (63 to 70 DAS) declined with manure storage times from 0.27, 0.19 to 0.16, in $M_{4,}$ M_{12} M_{13}, respectively. As the season progressed (70 to 100 DAS), the NEV for DM improved to 0.33, 0.26 and 0.22 in $M_{4,}$ M_{12} $M_{13,}$ respectively. The NEV for N uptake was slightly less that DM production for all manures and sampling times though followed a similar trend (Table 9).

DISCUSSION

Nitrogen response curve

The growth of canola showed a positive linear response to increasing rates of inorganic-N fertilizer up to 200 kg N ha^{-1} throughout the growing seasons (Figure 1).The significant increase in N uptake with increasing inorganic-N fertilizer indicated that mineral N was readily taken up by plants ($P<0.05$; Tables5 and 6). Concentrations of shoot N were below the critical concentration required for maximum yield (Reuter and Robinson, 1997) for all treatments throughout the experiment. This indicated that N was the limiting factor for plant growth on this infertile site as all other basal nutrients were provided. The low rainfall may have reduced potential yield over both years during the experiment, though still enabled the NEV in the manure to be calculated relative to inorganic-N.

Mineralisation of organic nitrogen in cattle manure

The application of inorganic-N resulted in higher yields compared to all sources of stockpiled cattle manure-N (M4, M12 and M13) at equivalent rates of application ($P<0.001$). Cattle manure typically comprises over 97% of total N in the organic fraction, which is slowly mineralized to inorganic N before it is available to plants (Larney and Angers, 2012). The design of the experiment accounted for the high solubility of inorganic-N fertilizer (urea) which was applied as a split application over the growing season to prevent N loss, whereas cattle manure was applied as a single application. The supply of N from the stockpiled manure treatments compared to inorganic N fertilizer showed that cattle manure was not adequate to supply the N requirements for plant production in the short term owing to the low rate of mineralization where soil fertility was low.

Nitrogen equivalent values(NEV) of cattle manure compared to inorganic fertilizer

At all N rates and sampling dates, the percentage NEV of the three cattle manure treatments compared to inorganic N fertilizer ranged from between 16 and 33% for DM production and between11 and 28% for N uptake, with lower values noted as the storage time of the manure increased (Table 9).Other researchers have shown a wide variation in NEV up to 59% for cattle manure in comparison to inorganic-N fertilizer in the first year of application due to factors such as the quality of the manure (the species, age and production level of the animal and diet), housing, manure collection and manure storage systems; further influenced by crop type and soil properties (Table 1). For example, manure applied as slurry or to irrigated crops may have a higher NEV, for example, values of 40 to 70% reported by Zhang et al. (1998)in the first year of application to irrigated corn. In cattle on better diets, the value can be higher also, for example NEV from 40 to 73% reported by Rigby (2008).The low N content of manures used in our study and low NEV reasonably reflects the poor quality diets of cattle in SSA and poor storage conditions of the manure compared to studies elsewhere. Duan et al. (2016) suggest that animal manure when managed and utilized appropriately can replace 70% of nitrogen (N) demand met through inorganic fertilizers in agriculture. It has long been established that the mineralization of N is generally greater for fresh cattle manure compared with manure stored for longer (Paul and Beauchamp, 1994).

Changes in nitrogen equivalent values (NEV) over the growing season

The release of available N from organic compounds during manure decomposition is very gradual and, in a manure-based cropping system, mineralization continues until after harvest (Mallory et al., 2010; Sousa et al., 2016). Much of this is determined by factors such as organic composition of the residue, soil temperature and water content, drying and rewetting events, and soil characteristics. In our study, the NEV of cattle manure increased over the growing season from 15 to 21% for each measure of crop response. The results indicate that on average approximately five times more total N equivalent would be required in manure to obtain comparative plant growth where all other nutrients are not limiting. The NEV of manure compared to inorganic-N may increase further pastthe100 DAS used in this study. It has been shown that longer-term accumulated applications of cattle manure release a sufficient supply of N each year to meet crop nutrient requirements without the annual addition of inorganic fertilizers (DeLuca and DeLuca, 1997; Helgason et al., 2007).

For each sampling date, NEV for manure compared to inorganic N was higher for DM production than for N uptake over both sampling times (Table 9). This would tend to indicate that factors in the manure other than N, such as other nutrients, organic matter and soil chemistry contributed to plant growth. For example, a neutral soil pH preferred by plants is often maintained by cattle manure (Benke et al., 2008). Although soil pH and other parameters were not tested in this study, they may have influenced plant growth. As this study was conducted in a dryland temperate environment, the N mineralization rates may differ from tropical conditions where soil properties, soil moisture and temperatures may be different and needs to be confirmed.

Effect of storage on nitrogen content in manure

In this study, increasing the storage time of cattle manure reduced its ability to supply N to the plant. The concentration of total N in manure ranged from 1.31% at 4 months (M_4) storage to 0.32% at 13 months (M_{13}) storage. In comparison, fresh cattle manure typically has a higher N content, for example 2.1% (Pettygrove et al., 2009) and up to 6.2% total N in unstabilized livestock waste in the UK (Rigby,2008). The concentration of N in manure used in our study was lower than the range of manures used elsewhere, but indicative of cattle manures in SSA.

In our study, cattle manure stored for 4 months (M_4) had a mean NEV of 30% for DM production and 19% for N uptake compared to inorganic N over two sampling dates. As the storage time of the manure increased to 13 months (M_{13}), the mean NEV reduced to 19% for DM production and 15.5% for N uptake compared to inorganic N over two sampling dates (Table 9). Gale et al. (2006) and Heinrich (2009) found that manure and other organic materials could be grouped into categories with similar N mineralization rates based on how they were produced, treated, and stored. This parameter is a very important determinant of the fertilizer value or nutrient use efficiency of the manure after its field application (Rashid et al., 2013, 2017a; Shah et al., 2016, 2018). Generally, stockpiling is not that effective in maintaining N in manure; for example, Shah et al. (2016) found that N recovery by maize for various treatments was as follows; fermented manure (39% of the field applied N) followed by the manure end products obtained from storing it in roofed building (31%), stockpiled (29%) or turned during storage (20%). In stockpiled manure, losses of ammonium will occur reducing the total N content and rainfall will further reduce N content due to the leaching of nitrate. Losses of total N occur following handling and transport which release ammonia to the atmosphere with good storage necessary to retain the fertilizer value of manure and improve crop yield (Holmes, 2007; Gooch and Wedel, 2010). High NEV, up to 70% has been reported when manure is stored for shorter periods of time (Powell et al., 2010).The application of freshly excreted manures incorporated immediately into the soil can improve the nutrient value of the soil (Gachimbi et al., 2009). However, some storage time of manure is required as the ammonia in fresh manure can burn crops if applied directly to the crops (Muhereza et al., 2014). The spreading of manure can result in the loss of ammonia (NH_3), especially during warm, dry weather (Stelt et al., 2007).

Rates of cattle manure required to optimize plant growth

Compared to inorganic fertilizers, higher volumes of manure must be applied for equivalent plant growth. For example, in order to supply 100kg N ha, approximately 25dry t ha of aged (4 months) cattle manure (~1.3% total N) would be required. The large volumes of manure required would be difficult to source and apply and may not be feasible for smallholder farmers. As the total N content of the manure decreases with storage (that is, ~0.32% total N), the higher volumes required are not practical to apply. Application rates of cattle manure could be reduced by improving the quality of manure through better storage to reduce the loss of N. Storage could be improved using covered shelters and impermeable floors to better retain N prior to spreading.

Organic recycling practices such as mulching, composting and manuring are well known in SSA and may help to reduce N loss (Zake et al., 2005; Gichangi et al., 2006; FAO, 2019).

Barker (2010) reported that cattle weighing 454 kg body weight produce 26 kg of faeces of fresh manure per day, with a total annual production including bedding of 16-18 tonnes of manure. An application of 49,421 kg $ha^{-1}$of good quality wet manure per year (that is,5 cattle) would supply about 225 kg of total N. Hence, 49 t wet manure ha^{-1} is often referred to as the agronomic rate of application, with N application based on the amount required by crops. From this application, about 45 kg N has been suggested as potentially available (Barker, 2010). To put this into perspective, manure from a total of one to two cattle would be required to fertilize a hectare of crop in SSA to provide the N equivalent of 100 kg ha^{-1}. However, the body weight of cattle in SSA is smaller than the Barker (2010) study due to lower quality diets including limited pastures and no supplementary feeds and compounded by lack of improved breeds. Hence in SSA, the use of cattle manure as a fertilizer is constrained by low manure output and quality.

Other options to improve crop N nutrition

Other options to improve crop N nutrition in SSA should be explored and could include supplementation of cattle manure with inorganic-N fertilizer. Nyamangara et al. (2005) concluded that smallholder farmers in Zimbabwe and similar SSA countries could exploit the combined application of manure and N fertilizer to increase maize yield. Opportunities exist to incorporate more effective crop rotations with legume crops to make better use of N_2 fixation by rhizobia and beneficial microorganisms and could reduce the amount of synthetic N fertilizer needed to maintain the N balance. The release of N bound in crop residues contributes to overall N fertility (Danga et al., 2009; Tonitto and Ricker-Gilbert, 2016). In addition to an overall increase in soil N by legumes, the benefits to healthy farming systems from crop rotations have been well established (St. Luce et al., 2016; Grant et al., 2016; Bainard et al., 2017). Benefits include effective weed control, breaking the persistence of soil-borne pathogens, increasing crop tolerance to abiotic stress, improved soil properties and improved soil microbial lbiodiversity. Overall, a diverse rotation system will enhance the long-term resilience of the farm system (Abraham et al., 2014; Thiessenet al., 2015). In order to improve adoption of a legume rotation systems approach at the local farm level, relevant economic and agro-environmental policies are required in SSA.

Conclusion

The N equivalent value of cattle manure compared to inorganic-N fertilizer was dependent on its N content and hence analysis of the N content in cattle manure prior to use is essential for calculating agronomic application rates. The storage of cattle manure from 4 months to 13 months decreased the total N content from 1.3 to 0.32% and reduced N available for plant growth; therefore, better storage conditions of cattle manure are needed to reduce N loss over time. The N equivalent value of cattle manure stored for 4 months (1.3% total N content) was approximately 30% for DM production and 19% for N uptake over the growing period compared to inorganic-N fertilizer and reduced significantly when stored for up to 13 months.The volumes of cattle manure required to satisfy the N fertilizer recommendations compared to inorganic fertilizer may be impractical and further options need to be explored, such as supplementation with inorganic fertilizer or the inclusion of crop rotations with legume crops.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Abraham B, Araya H, Berhe T, Edwards S, Gujja B, KhadkaRB,Verma A (2014). The system of crop intensification: Reports from the field on improving agricultural production, food security, and resilience to climate change for multiple crops. Agriculture and Food Security 3:4-16.

Bainard LD, Navarro-Borrell A, Hamel C, Braun K, Hanson K, Gan Y (2017). Increasing the frequency of pulses in crop rotations reduces soil fungal diversity and increases the proportion of fungal pathotrophs in a semiarid agroecosystem. Agriculture, Ecosystems and Environment 240:206-214.

Barker AV (2010).Science and Technology of Organic Farming. New York, USA: CRC Press, Taylor and Francis Group, LLC.

Benke M, Hao X, Chang C (2008). Effects of long-term cattle manure applications on soil, water and crops: Implications for animal and human health In "Development and Uses of Biofortified Agricultural Products" edited by G. Banuelos and Z.-Q. Lin CRC Press, Boca Raton, FL. (in press).

Chambers BJ, Lord EI, Nicholson FA, Smith KA (1999). Predicting Nitrogen Availability and Losses Following Application of Organic Manures to Arable Land: Manner. Soil Use and Management 15(3):37-143.

Croppenstedt A, Demeke M, Meschi MM (2003). Technology adoption in the presence of constraints: the case of fertilizer demand in Ethiopia. Review of Development Economics 7(1):58-70.

Danga BO, Ouma JP, Wakindiki IIC, Bar-Tal A(2009). Legume-wheat rotation effects on residual soil moisture, nitrogen and wheat yield in tropical regions. Advances in Agronomy 101:315-349.

DeLuca TH, De Luca DK (1997). Composting for Feedlot Manure

Management and Soil Quality. Journal of Production Agriculture 10:235-241.
Duan Y, Xu M, Gao S, Liu H, Huang S, Wang B (2016). Long-term incorporation of manure with chemical fertilizers reduced total nitrogen loss in rain-fed cropping systems. Science Report 6:33611.
FAO (2019).Nitrogen inputs to agricultural soils from livestock manure: New statistics. In Integrated Crop Management, P. 24.
Esfahani A, Luo Y, Li C, Benke MB, Hao X, Larney FJ (2016). Yield and nutrient uptake of barley and camelina amended with manure from cattle fed barley, triticale dried distillers grains with solubles, and flaxseed diets. Canadian Journal of Soil Science 97(2):99-108.
Ferguson BR, Nienaber JA, Eigenberg RA, Woodbury BL(2005).Long-Term Effects of Sustained Beef Feedlot Manure Application on Soil Nutrients, Corn Silage Yield, and Nutrient Uptake. Journal of Environmental Quality 34:1672-1681.
Gachimbi L, Zake J, Ebanyat P, Ergano K, Abduke M, van Keulen H(2009). Cattle Manure Management in East Africa: Review of Manure Quality and Nutrient Losses and Scenarios for Cattle and Manure Management. Report 258.Lelystad: Wageningen UR Livestock Research, Partner in Livestock Innovations.
Gale ES, Sullivan DM,CoggerCG, BaryAI, Hemphill DD,MyhreEA(2006).Estimating Plant Available Nitrogen Release from Manures, Composts, and Specialty Products. Journal of Environmental Quality 35(6):2321-2332.
Gichangi EM, KaranjaNK,Wood CW(2006). "Composting Cattle Manure from Zero- Grazing Systems with Agro-Organic Waste to Minimize Nitrogen Losses in Smallholder Farms in Kenya.Tropical and Subtropical Agro-ecosystems 6:57-64.
Gooch C, WedelAW(2010). Effective Means of Handling Sand-Laden Dairy Manure.http://www.ansci.cornell.edu/prodairy/index.html.
Grant CA, O'Donovan JT, Blackshaw RE, Harker KN, Johnson EN, Gan Y,Ramnarine R (2016). Residual effects of preceding crops and nitrogen fertilizer on yield, crop, and soil N dynamics of spring wheat and canola in varyingenvironmentson the Canadian prairies. Field Crops Research 192:86-102.
Griffin TS, Giberson E, Wiedenheft M (2002).Yield Response of Long-Term Mixed Grassland Awards and Nutrient Cycling under Different Nutrient Sources and Management Regimes. Grass and Forage Science 57:268-278.
Heinrich A (2009).Nitrogen fertilizer value of solid and liquid dairy wastes produced in California's San Joaquin Valley". M.S. diss., University of California, Davis. In Dissertations and Theses: Available from http://www.proquest.com (publication number AAT 1468085; accessed October 20, 2011).
Helgason BL, Larney FJ, Janzen HH, Olson BM(2007). Nitrogen Dynamics in Soil Amended with Composted Cattle Manure. Canadian Journal of Soil Science 87:43-50.
Holmes BJ (2007). Sand Laden Manure Handling Alternatives Biological Systems Madison, Engineering Department, University of Wisconsin.
Kaizzi KC, Mohammed MB, Nouri M (2017). Fertilizer Use Optimization: Principles and Approach. In: Fertilizer Use Optimization in Sub-Saharan Africa. Charles S. Wortmann and Keith Sones (eds). CAB International, Nairobi, Kenya, pp. 9-19.
Katuromunda S, Sabiiti EN, Bekunda AM (2012). Effect of method of storing cattle faeces on the physical and chemical characteristics of the resultant composted cattle manure. Uganda Journal of Agricultural Sciences 13 (2):95-106.
Kazemeini SA, Hamzehzarghani H, Edalat M (2010). The Impact of Nitrogen and Organic Matter on Winter Canola Seed Yield and Yield Components.Crop Production and Plant Breeding Department, College of Agriculture, Shiraz University, Shiraz, Iran. Australian Journal of Crop Science 4(5):335-342.
Larney FJ, Angers DA (2012). The role of organic amendments in soil reclamation: A review.Canadian Journal of Soil Science 92:19-38.
Leip A, Ledgard S, Uwizeye A, Palhares JCP, Aller MF, Amon B, Binder M, Cordovil CMDS, De Camillis C, Dong H, Fusi A, Helin J, Hörtenhuber S, Hristov AN, Koelsch R, Liu C,Masso C, Nkongolo NV, PatraAK, Redding MR, Rufino MC, Sakrabani R, Thoma G, Vertès F,Wang Y (2019). The value of manure-Manure as co-product in life cycle assessment. Journal of Environment Management 241:293-304.
Mallory EB, Griffin TS, Porter GA (2010).Seasonal Nitrogen Availability from Current and Past Applications of Manure. Nutrient Cycling in Agroecosystems 88(3):351-360.
Muhereza I(2011).The Application of Cattle Manure to Improve Soil Fertility for Crop Production in Uganda. PhD Thesis, Muresk Institute of Agriculture, Curtin University of Technology, Perth, Western Australia.
Muhereza I, Pritchard D, Murray-Prior R (2014).Utilisation of Cattle Manure and Inorganic Fertilizer for Food Production in Central Uganda. Journal of Agriculture and Environment for International Development 108(2):135-151.
Mwangi W (1996). Low Use of Fertilizers and Low Productivity in Sub-Saharan Africa.NRG Paper 96-05. Mexico, D.F.: CIMMYT.
Ndambi OA, Pelster DE, Owino JO, de Buisonjé F, Vellinga T (2019). Manure Management Practices and Policies in Sub-Saharan Africa: Implications on Manure Quality as a Fertilizer. Frontiers in Sustainable Food Systems 3:29. URL=https://www.frontiersin.org/article/10.3389/fsufs.2019.00029.
Nyamangara J, Mudhara M, Giller KE (2005).Effectiveness of cattle manure and nitrogen fertilizer application on the agronomic and economic performance of maize. South African Journal of Plant and Soil 22(1):59-63
Paul JW, Beauchamp EG (1994). Short-Term Nitrogen Dynamics in Soil Amended with Fresh and Composted Cattle Manures. Canadian Journal of Soil Science 74:147-155.
Payne RW, Harding SA, Murray DA, Soutar DM, Baird DB, Glaser AI, Welham A, Gilmour RR, Thompson B, and Webster R(2011). Introduction to Genstat for Windows, 14th Edition: VSN International, Hemel, Hempstead, Hertfordshire HP1 1ES, UK.
Pettygrove GS, Davis A, Heinrich L(2009).Dairy Manure Nutrient Content and Forms.University of California Cooperative Extension Manure Technical Bulletin Series. http://manuremanagement.ucdavis.edu.
Powell JM, Gourley CJP, Rotz CA, Weaver DM (2010). Nitrogen use efficiency: a potential performance indicator and policy tool for dairy farms. Environmental Science Policy13:217-228.
Rashid MI, de Goede RG, Brussaard L, Lantinga EA (2013). Home field advantage of cattle manure decomposition affects the apparent nitrogen recovery in production grasslands. Journal of Soil Biology and Biochemistry 57:320-326.
Rashid MI, Lantinga EA, Brussaard L, de Goede RGM (2017a). The chemical convergence and decomposer control hypotheses explain solid cattle manure decomposition in production grasslands. Journal of Applied Soil Ecology 113:107-116.
Reijs JW, Soneld MPW, Sørensen P, Schils RLM, Groot JCJ, Lantinga EA(2007). "Effects of Different Diets on Utilization of Nitrogen from Cattle Slurry Applied to Grassland on a Sandy Soil in the Netherlands. Agriculture, Ecosystems and Environment 118:65-79.
Reuter DJ, Robinson JB (1997). Plant Analysis: An Interpretation Manual. Collingwood, Vic: CSIRO Publishing.
Rigby H (2008). "Quantifying the Agronomic Value of Biosolids and Biowastes."PhD Thesis.Department of Chemistry, Imperial College, London, UK.
Shah GM, Shah GA, Groot JCJ, Oenema O, Raza AS, Lantinga EA (2016). Effect of storage conditions on losses and crop utilization of nitrogen from solid cattle manure. Journal of AgriculturalScience 154:58-71.
Shah GA, Shah GM, Rashid MI, Groot JC, Traore B, Lantinga EA (2018). Bedding additives reduce ammonia emission and improve crop N uptake after soil application of solid cattle manure. Journal of Environment Management 209:195-204.
Sheahan M, Barrett CB (2014). Understanding the agricultural input landscape in Sub-Saharan Africa: recent plot, household, and community-level evidence. World Bank Policy Research Working Paper, 7014.
Snijders P, van der MH, Onduru D, Ebanyat P, Ergano K, Zake J, Wouters B, Gachimbi L, van Keulen H (2013). Effects of cattle and manure management on the nutrient economy of mixed farms in East Africa: A scenario study. African Journal of Agricultural Research 8(41):5129-5148, 24.
Stelt B, Temminghoff E, Petra VV, Riemsdijk W (2007). Volatilization

of ammonia from manure as affected by manure additives, temperature and mixing. Journal of Bio resource Technology 98:3449-55.
Sousa JR, Cabral F, Coutinho J (2016). Assessment of N Mineralization and N Leaching in Soil Using a New in situ Incubation Method. Communications in Soil Science and Plant Analysis 47(19):2157-2167.
Stewart ZP, Pierzynski GM, Middendorf BJ, Prasad VPV (2020). Approaches to improve soil fertility in sub-Saharan Africa. Journal of Experimental Botany 71(2):632-641, https://doi.org/10.1093/jxb/erz446.
St. Luce M, Grant CA., Ziadi N, Zebarth BJ, O'Donovan JT, Blackshaw RE, McLaren DL(2016). Preceding crops and nitrogen fertilization influence soil nitrogen cycling in no-till canola and wheat cropping systems. Field Crops Research 191:20-32.
Thiessen MJR, Entz MH, Wonneck MD (2015). Review: Redesigning Canadian Prairie Cropping Systems for Profitability, Sustainability, and Resilience. Canadian Journal of Plant Science 95:1049-1072.
Tonitto C, Ricker-Gilbert JE (2016). Nutrient management in African sorghum cropping systems: Applying meta-analysis to assess yield and profitability. Agronomy for Sustainable Development 36:1-19.
Whitehead DC (1995). Grassland Nitrogen Oxon, UK: CAB International.
Zake J, Tenywa JS, Kabi F (2005). Enhancement of Manure Utilization in Smallholder Cattle Management Systems for Crop Production in Central Uganda. African Crop Science Conference Proceedings 7:1067-1071.
Zhang H, Smeal D, Tomko J(1998). Nitrogen Fertilisation Value of Feedlot Manure for Irrigated Corn Production. Journal of Plant Nutrition 21:287-296.

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Role of farmers' organizations (FOs) in the strengthening of the technical and organizational capacities of farmers in Mezam Division of the North West Region of Cameroon

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This study was carried out from January 2018 to March 2019 in Mezam Division of the North West Region of Cameroon on the role of farmers organization (FOs) in the strengthening of the technical and organizational capacities of farmers, particularly the case of the Program for the Improvement of Competiveness of Family Agro-pastoral Farms (PCP-ACEFA) and the North West Farmers' Organization (NOWEFOR). The objective of the study was to analyze the role of FOs on the technical and organizational strengthening of the capacities of farmers and the organization in Mezam Division of the North West Region of Cameroon. Secondary sources data were reviewed. Primary source data were obtained directly from the field. Two hundred and eighty (280) farmer members of the farmers' organisations were interviewed using a semi-structured questionnaire and 7 leaders were interviewed using an interview guide. In addition, direct observations were made. The data obtained were analyzed using SPSS. The findings showed that the contribution on the development of technical and organisational capacities of the farmers was overall positive as farmers had improved skills in input supply (51.42%), production (38.57%), and market access (27.14%) compared to non-beneficiaries. The contribution on the strengthening of the organisation as a whole was overall positive since it had permitted FOs to respectively employ technical staff (52.85%) and boosted membership (45.71%) and improved group input supplies and group sales in the organisation. This study concluded that farmers' organizations are pivotal in the strengthening of the technical and organisational capacities of farmers and their organisations.

Key words: Contribution, farmers, farmers organisation, technical and organisational**,** rural development.

INTRODUCTION

Farmers' Organizations (FOs) emerged in the world due to farmer-felt needs such as sharing of local resources

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and inputs (seeds, breeds, know-how and experience) and market pressures such as prices and access to markets (Msuta and Urassa, 2015:2343). NOWEFOR (2017:2) signaled that farmers organizations play a pivotal role in providing training, sharing of experiences on production and marketing techniques; organization of wholesale purchases of inputs and group sales of farm produce. Gouët et al. (2009:75) highlighted that farmers organizations (FOs) promote rural development by serving as a corridor for sharing information, co-coordinating activities, making collective decisions, creating opportunities for producers to get more involved in value-added activities such as input supply, credit, processing, marketing and distribution on the one hand and create awareness in view of defending farmers interest on the other hand.

In the North West Region of Cameroon, FOss have services such as trainings in agriculture to members, to boost income and well-being at large. Fongang and Fru Mbangari (2017:3) reported that the farmers targeted in order to improve their living conditions through capacity building and training in production and group marketing, appear not to have been empowered in such a manner that will guarantee the sustainability of the farmers' movements. Besides, several studies have been carried out on the evaluation of farmers organizations (Benoit, 2006:15; NOWEFOR, 2012:25) but it appears no impact assessment has been carried out at the individual and organizational levels to show whether the role of strengthening the capacities of by FOs have a positive impact on the farmers. It is for this reason that this study was undertaken to determine the technical and organizational role which have been brought about by FOs on the target population at the levels of the individual and organizational within the framework of poverty alleviation. The objective of the study was to analyze the role of FOs on the technical and organizational strengthening of the capacities of farmers and the organization in Mezam Division of the North West Region of Cameroon.

MATERIALS AND METHODS

Study area

The study was carried out in Mezam Division of the North West Region. Mezam Division is located between latitudes 5°40' and 7°50'North and longitudes 9°80' and 11°51' east of the Greenwish Meridian (https://en.wikipedia.ord/wiki/Mezam.). Mezam has a total surface area of 1,841.45 km^2 with a total population of 524,127 inhabitants in the 2005 census. The agricultural population is estimated at 258467 inhabitants representing 43.07% of farm families (Republic of Cameroon, 2015). This population belongs to a large set of Ethnic groups, made up of several tribes such Ngemba (Awings, Mankons, Bafuts, Nkwens, Pignins, Akums and Njongs), Mugahkah (Bali), Bei (Baba IIs, Bafochus), etc. (Figure 1). The climate is of the tropical savannah type with two distinct seasons: The rainy and the dry seasons. The rainy season starts from mid-March to mid-October. The dry season is characterized by winds and runs from late October to mid-March. Vegetation comprise doted parches, artificial and natural forest, short and thick grasses, hence its name "Grass-field.

Data collection

A descriptive and cross-sectional research design was used to generate data for this study. Data for the study were obtained from two sources: Data from secondary and primary sources. Secondary source data were obtained from relevant literature existing in documents and archives of several structures such as: The central library of the University of Dschang, British Council library in Bamenda, DDARD annual reports, ACEFA activity reports, project reports, evaluation reports and from the internet, etc. In order to characterize these FOs and analyze their activities, secondary source data from DDARD annual reports, ACEFA activity reports, project reports, evaluation reports, baseline studies reports, mission reports and additional information from administrative authorities were used. The information were summarized such as to bring out a clear picture of the type of FOs operating in the region on the one hand and analyses of its partners on the other hand. Primary source data were obtained via observations, interviews (focus group discussions, meetings) and the administration of questionnaires to the beneficiary farmers covered by the FOs.

Sampling

A stratified random sampling method was used. The population of the study is divided into strata. Firstly, out of the five Divisions, Mezam Division was chosen because it has the highest number of FOs constituting 41% of the 16425 FOs in the North West Region. Secondly, 1% of the 6725 FOs in Mezam division of the NWR was obtained to constitute the sample size which gave us 70 FOs. Reasoning being that the 6725 FOs was information from the Regional Delegation of Agriculture and Rural Development, but as we went to the field, it was noticed that the information gotten from PCP-ACEFA and NOWEFOR in Mezam, based on accessibility and security was only 403 FOs as shown in Table 1. As such 17% of the 403 FOs in Mezam were obtained to constitute the sample size which gave us 70 FOs. Thirdly, for comparison purposes and following aid intervention, the sample size was also broken down into 40 beneficiary FOs and 30 non beneficiary FOs. Fourthly, Four (04) members belonging to each of the farmers' organisations in the seven Subdivisions' of the aid in Mezam Division were interviewed. These data obtained were analysed using Statistical Package for Social Sciences (SPSS). The non-descriptive statistical tools were used to analyze the findings. These findings are presented in form of simple cross-tables, frequencies distributions percentages and bar charts

Theoretical framework and concepts

Asante-Addo et al. (2016:1) reported that farmer organizations in Ghana contributes or play an important role in the granting of credit and its services to farmers, training them in their activities and increasing membership in their organization. Farmer organizations involve in credit programs because of improved loan access for farming purposes and savings mobilization. Such market smart strategies have the potential to improve farmers' access to timely credit and to reduce rural poverty. For Gouët et al. (2009:3) farmer organization are characterized based on their history, reason of existence, objectives, and ambits of actions, degree of formalization, and their domain of intervention.

All impact assessments embody three main elements: A model of the impact chain that the study is to examine; the specification of

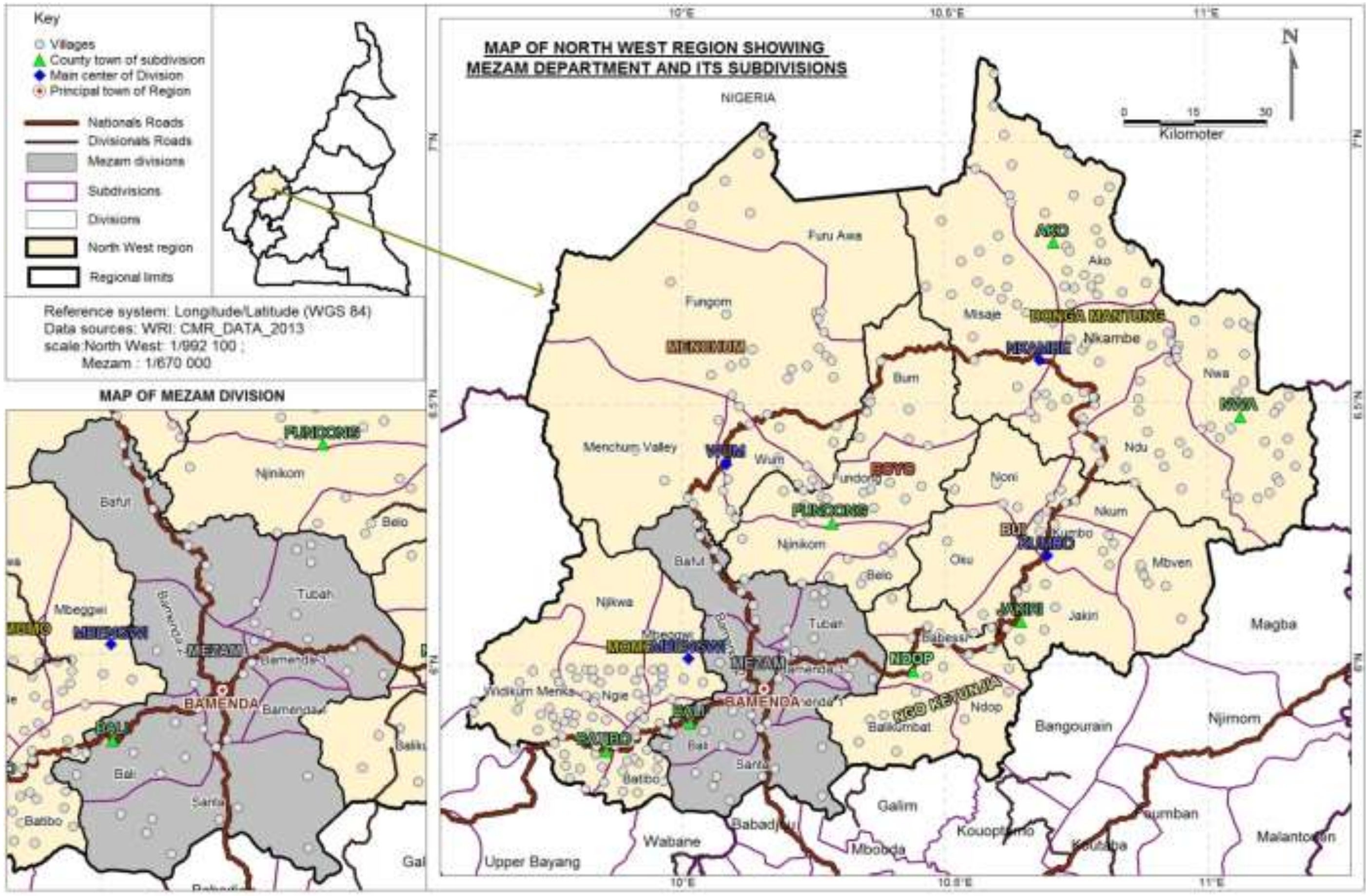

Figure 1. Map of the North West region showing Mezam division.
Source: World Research Institute, 2019.

Table 1. Treatment, control and differences before and after in impact assessment.

	Treatment	Control	Difference
Before	6	8	-2
After	12	10	**2**
Difference	**6**	**2**	**4**

Source: Bilal (2014).

unit(s) or levels, at which impact is assessed and the specification of the type of impact that are to be assessed. Impact Assessment (IAs) measure the difference in the key variables between the outcomes on “agents”(individuals, enterprises, household, community, etc.), which have experienced an intervention against the values of those variables that would have occurred had there been no intervention aid program (Hulme, 1997). Masud and Yontcheva (2005) measured the impact of external aid on Human Development indicators such as infant mortality and illiteracy using regression and the findings revealed that increased health expenditure per capita reduces infant mortality as does greater NGO aid per capita. In order to conduct a valid impact assessment, researchers first need to define their overall strategy which sets the course for the rest of the research process (Hulme, 1997; Koehler et al. 2007). Another non-experimental methods of impact assessment as agreed upon by the World Bank is the difference-in-differences and this method relies on key assumptions. For instance difference #1 compare over time, the situation before and after the program whereas difference #2 compare the treatment and control groups so as to measure changes between the outcomes on individuals, organizations, communities, etc. He argued that impact assessment is better achieved when the before-after and with-without approaches are combined to an overall treatment effect (Bilal, 2014) as indicated in Table 1.

Alternatively the study sought the indicators of role of FOs on the technical and organizational strengthening of the capacities of

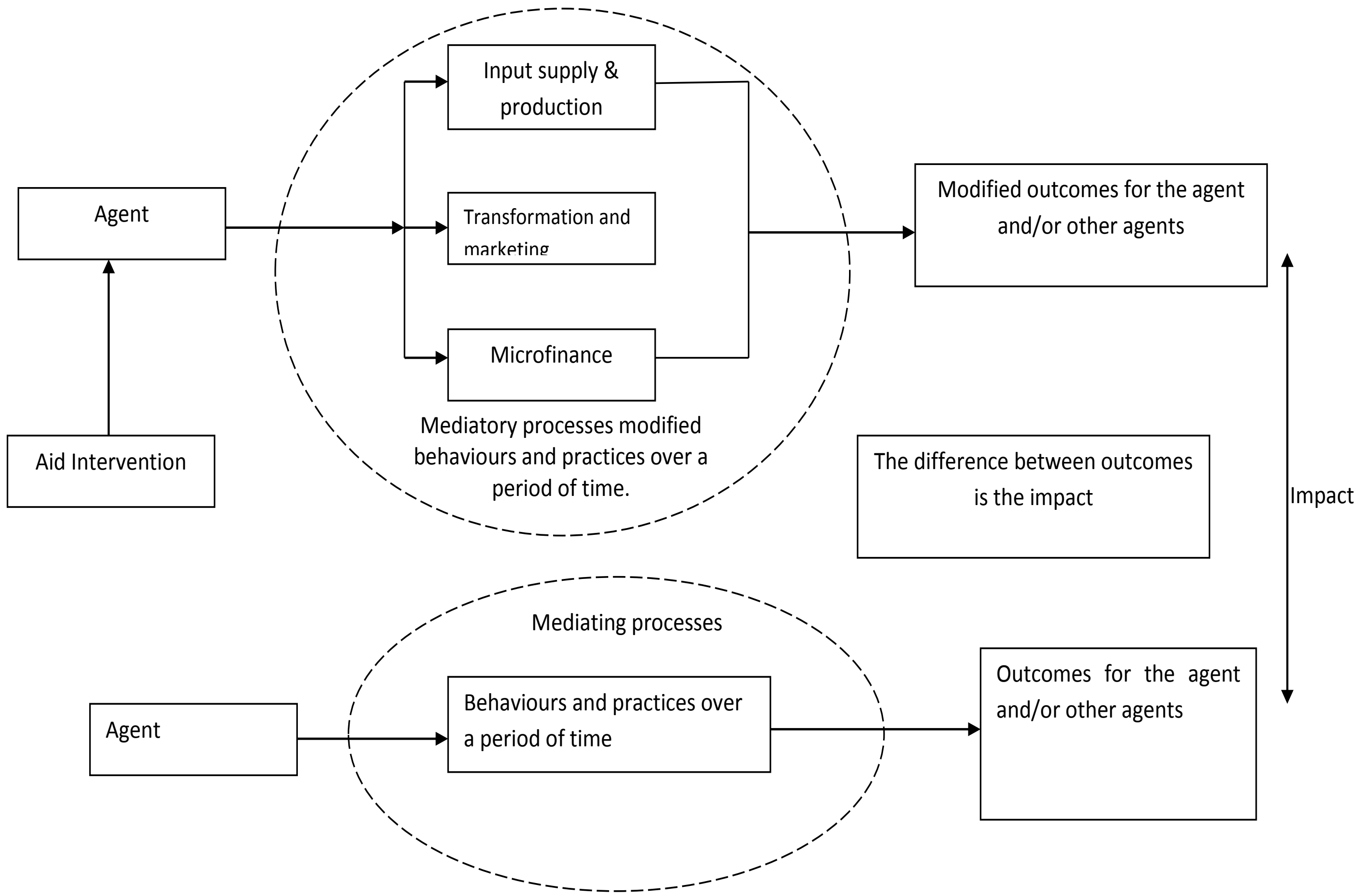

Figure 2. The operational model of the Impact chain for the study.
Source: Adapted from Hulme (1997:26).

farmers and the organization in the Region through an impact assessment of the observable and measurable changes between the outcomes on “agent” (individuals and organization) that have experienced aid interventions against the values of those variables that would have occurred had there been no aid intervention as shown in Figure 2. The findings will help concerned policy makers (PCP-ACEFA, SOS Faim Luxembourg and European Union) to take appropriate decisions in formulating aid assistance strategies that will help improve the living conditions of farmers and FOs.

RESULTS AND DISCUSSION

Socio economic characteristics of respondents

The main characteristics concerned here are sex, age, marital status, education and main income generating activities illustrated in Table 2.

As revealed by Table 2, women generally constitute 52.85% and men constitute 47.14% of the total respondents mean while the fraction of women beneficiaries stands at 27.14%. The percentage of women beneficiaries (27.14%) could be explained by the fact that one of the priorities of FOs was for their contribution to reach out to more women who were considered as the marginalized group in the division. The predominance of men in crop and livestock production as observed in this study is in agreement with the findings of Defang et al. (2014) who report that pig production is dominated by men in the urban and peri urban zones in Dschang-West region Cameroon.

Overall, 71.42% of the total respondents were between the age group 25 and 45 years. The mean age of the respondents was 40 years (± 5) indicating that a high proportion of the middle age respondents were involved in production. This is in line with the findings of Defang et al. (2014) who signal that majority of the adult (middle age) population of the society are involved in livestock production. A fraction of the active rural population of this division found between 25 and 45 years is therefore looked upon as the initiators of the development of crop and livestock production. Thirty percent (30.00%) of the respondents are aged 55 years. This increased in number of the old population could be explained by the

Table 2. Distribution of respondents by sex, age group, marital status, level of education and main income generating activity.

Parameters and characteristics	Category of beneficiaries		
	Beneficiaries (%)	Non beneficiaries (%)	Total [280(%)]
Sex			
Male	84 (30%)	64 (22.85%)	148(52.85%)
Female	76(27.14%)	56 (20%)	132(47.14%)
Age groups			
25-35	56(20%)	96(34.28%)	152(54.28%)
36-45	32(11.42%)	16(5.71%)	48(17.14%)
46-55	20(7.14%)	12(4.28%)	32(11.42%)
>55	52(18.57%)	4(1.42%)	56(20%)
Marital status			
Single	16 (5.71%)	36(12.85%)	52(18.57%)
Married	128(45.71%)	76(27.14%)	204(72.85%)
Widow(er)	16(5.71%)	0(0%)	16(5.71%)
Divorced	0(0%)	8(2.85%)	8(2.85%)
Level of Education			
Primary	28(10%)	24(8.57%)	52(18.57%)
Secondary	8(2.85%)	24(8.57%)	34(12.14%)
2nd cycle secondary	52(18.57%)	72(25.71%)	124(44.28%)
Higher	32(11.42%)	0(0%)	32(11.42%)
Main income generating activity			
Market gardening	32(11.42%)	20(7.14%)	52(18.57%)
Broilers	64(22.85%)	40(14.28%)	104(37.14%)
Piggery	72(25.71%)	52(18.57%)	124(44.28%)

fact that they were already based in the rural areas. Eleven percent (11.42%) of the respondents were between the age group 45 to 55 years.

As revealed by Table 2, 72.85% of the respondents were married and among them 45.71% were aid beneficiaries. Eighteen percent (18.57%) of the respondents are single and only 5.71% of them are beneficiaries. This 18% of the respondents who were single appears to be those who were found between the age group 15 to 35 years. This could be explained by the fact that they do not have responsibilities and access to land for farming. The high percentage of married respondents in the study zone agree with the results of Defang et al. (2014) and Fotso et al. (2014) in the West Region of Cameroon who highlighted that majority of the adult population of a society consist of married people. The implication of this is that housewives were still predominantly used as family labour for light farm operations.

As shown on Table 2, 100% of the respondents have a level of education comprising between primary, secondary and higher schools and 42.85% of them are beneficiaries. The high rate of the respondents in this study who had formal education agrees with the findings of Defang et al. (2014) who reported that a higher percentage of pig farmers in the urban and peri - urban zone of Dschang - West Region of Cameroon had formal education. Education may be of assistance to extension and FOs staff for easy communication and understanding of extension message, especially for application of new technology in swine production and management. The fact that 100% of them are literate could facilitate trainings, extension, advice, monitoring and evaluation. The implication is that literate farmers might be more likely to adopt good farming and health-management practices.

As shown in Table 2, 18.57% of the respondents are involved in market gardening as their main source of income. They are mostly youths who are single and found between the age group15 to 35 years. This could be explained by the fact gardening requires much physical efforts and adequate technical know-how. Thirty seven percent (37.14%) of the respondents who are involved in broilers production are found between the age group 35 to 55 years. This could be explained by the fact that broiler production requires little physical efforts and is very profitable and also one of the conditions for farmers to received support in poultry was for them to have a poultry house. Forty four percent (44.28%) of the respondents who are aged 55 years and above and are

Table 3. Distribution of respondents by acquired skills and abilities in farm business.

Parameters and characteristics	Category of beneficiaries		
	Beneficiaries (%)	Non beneficiaries (%)	Total [280 (%)]
Input supply skills			
Increased	144 (51.42%)	24 (8.57%)	168 (60.00%)
Constant	8 (2.85%)	96 (34.28%)	104 (37.14%)
Decreased	8 (2.85%)	0 (0%)	8 (2.85%)
Improved production skills			
Increased	108 (38.57%)	32 (11.42%)	140 (50.00%)
Constant	28 (10.00%)	68 (24.28%)	96 (34.28%)
Decreased	24 (8.57%)	20 (7.14%)	44 (15.71%)
Improved marketing skills			
Increased	76 (27.14%)	32 (11.42%)	108 (38.57%)
Constant	28 (10.00%)	64 (22.85%)	92 (32.85%)
Decreased	56 (20.00%)	0 (0%)	56 (20.00%)

Table 4. Group marketing of produce (gardening, broilers and piggery) in NOWEFOR.

Period	Speculation	No. of group sales	Quantities sold	Average prices per unit (in FCFA)
2007	Gardening Broilers Pigs	40	2010 tons 50000 birds 800 pigs	140 per kilogram 3200 per chicken 65000 per average pig
2008	Gardening Broilers Pigs	76	2600 tons 70000 birds 1000 pigs	150 per kilogram 3500 per chicken 72000 per average pig
2009	Gardening Broilers Pigs	114	3500 tons 95000 birds 2000 pigs	175 per kilogram 3700 per chicken 76000 per average pig
2010	Gardening Broilers Pigs	225	4700 tons 110000 birds 2100 pigs	200 per kilogram 3800 per chicken 82000 per average pig
2011-2014	Gardening Broilers Pigs	950	5500 tons 150000 birds 4000 pigs	333 per kilogram 4000 per chicken 85000 per average pig
2015-2017	Gardening Broilers Pigs	970	6000 tons 140000 birds 45000 pigs	366 per kilogram 4000 per chicken 90000 per average pig

mostly involved in piggery. It is observed that the old are mostly involved in piggery because it requires little technical knowledge and physical efforts though not very profitable compared to poultry.

Contribution of FOs to technical and organizational capacities of farmers

The contribution of FOs on improved production skills, organization of group sales and input supplies are presented in Tables 3 and 4.

Contribution at the level of the farmer

As revealed in Table 3, 51.42% of input supply skills and abilities of the beneficiary farmers increased compared to 8.57% of the non-beneficiaries. Table 3 also reveals that 38.57% of improved production skills and abilities of the

Figure 3. Training of trainers' workshop at NOWEFOR**.**

Figure 4. Training of farmers in Mforyah on improved techniques on reduced chemical residues tomatoes production**.**

beneficiary farmers increased compared to 11.42% of the non-beneficiary farmers. Twenty seven percent (27.14%) of improved marketing skills and abilities of the beneficiary farmers increased compared to 11.42% of the non-beneficiary farmers. This could be explained by the fact that FOs has empowered their members or farmers through capacity building sessions and workshops so as to enable them to fully assume their roles and pilot their farm businesses. Figure 4 shows a training session conducted by FOs at Mforyah Integrated Farmers Union (MIFU). Also the mission of FOs is to improve the internal organization, strengthen the economic and financial capacities and service delivery to members. As agriculture is the main stay of the people, investing in its promotion and the strengthening of the capacities of farmers is important for poverty alleviation and farmers empowerment

The farmers were trained on input needs assessments skills, improved production techniques and marketing for viable commodity chains such as market gardening, poultry piggery, ginger, etc.. Usually before any training of farmers commenced, a training of trainers' workshop was organized that brought together farmer resources persons, agriculture and livestock officers of the Ministries of Agriculture and that of Livestock and local partner organizations to harmonize the training packages per subsector of production like market gardening, poultry and piggery. The farmers were trained on thematic areas such as joint inputs needs assessments, joint commands and negotiations for bulk inputs, group supplies and distribution as shown in Figure 3.

The following strategies were used to implement the training at the farmer level in and around Mezam Division.

Building farmers capacities: After more than ten years of collaboration between NOWEFOR and farmers both parties recognized that NOWEFOR had acquired

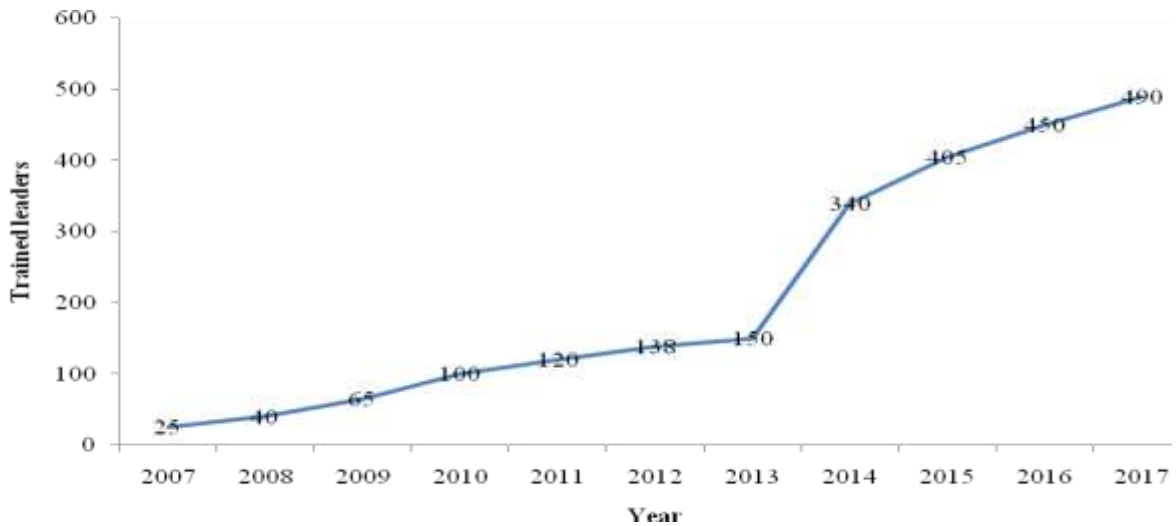

Figure 5. Evolution of trained farmers of NOWEFOR.

sufficient capacities to assume its organizational responsibilities. In 2008, NOWEFOR put in place a successful system of peer training which has been extended to all unions and farmers. This system consisted of identifying elite farmers and some who have capacities to organize trainings for other peer farmers. These resource persons collaborated with project staff to design training modules and technical slips and implemented the trainings. These training modules served as guides to farmer resource persons to continue offering training to other farmers after the workshop ends. The technical slips were distributed to farmers as reference during farm management. Participatory adult learning tools and skills were used to facilitate the trainings. This included brainstorming, group works, restitution, role plays and exchange visits.

The number of trained farmer resource persons or leaders increased with time from 25 leaders in 2007 to 490 leaders in 2017 as illustrated in Figure 5

Evolution of the contribution and targeting: The FOs followed a rolling approach in implementing the trainings. They proceeded from a nucleus of farmers who met up with the performance criteria as follows:

1. Have paid annual dues and all other levies in the union
2. Been registered in the credit house
3. Not be a delinquent member
4. Been a producer (have a current farm)
5. Been saving at least once a month in the credit house
6. Must have been in the union for at least 2 years
7. Must be resident in the Zone
8. Must not be on a permanent salary

The trainings sessions or workshops started up with more experienced farmers and peer training system was used, thus reducing training cost and at the same time built a strong success base for subsequent expansion. New farmers were therefore taken care of when they have also met their minimum performance criteria as it played a big role on material obligations and financial contributions.

Production sector approach: The commodity chain approach was used here to develop crop and livestock production. This entailed working on key areas along the value chain from production to marketing of products. In this approach farmers were organized around the main commodities; market gardening, poultry and pig production. This organization was needed to facilitate training of farmers and exchange of experiences amongst a critical number of farmers involved in a particular production chain. Once farmers were organized, group input need assessments, group commands and supplies of inputs were easily carried out as illustrated in Figure 6. To succeed in this approach, local management committees were elected and trained to handle the input supply mechanisms. This was needed to ensure the continuity of the actions by the local farmer leaders after the initial aid period. Group commands and better negotiation of prices developed the negotiation capacities of the individual farmers. It also enhanced economies of scale and gains in price reductions during the purchase on inputs. Because large quantities were commanded at a time, the transportation of the supplies was facilitated. Farmers had inputs on time in their communities. This approach also facilitated timeliness in farm activities and thus increased the productivity of the agricultural enterprises.

Integration of livestock and crops / promotion of organic manure: Crop and livestock integration was the

Figure 6. Group input supply of cement and chicks to boost poultry subsector in Bambui.

Figure 7. Farmers in Mfoyah receiving piglets and feed through FOs to boost pig subsector.

principle promoted. These farmers were trained amongst others to manage organic manure from their livestock farms and to integrate this in their gardens. NOWEFOR has experience in manure management and the production of low external input – chemical residue free market gardening crops. This focus on biological agriculture was needed to maintain the quality of the products and therefore fetch better prices for the farmers. The integration of organic manure also promoted soil fertility conservation and improvement. As the province is witnessing rapid fertility depletion, organic manure will help redress this situation and maintain high yields. The integration approach reduced future dependence of farmers on inorganic, expensive, scarce and environmentally unfriendly manure.

Direct production assistance: Farmers after receiving training on improved production skills per subsector were provided with necessary productive resources to scale up or start up their enterprises. The external input was combined to local dynamics and local contributions to realize the projects. And as such this partnership was needed for the appropriation of the actions. Inputs provided included seeds, piglets, chicks, manure, agrochemicals, etc. as shown in Figure 7. However, farmers were trained to control the use of these products in the expansion of their farms. Farmers were also trained on integrated pest management and product handling. All of these were to ensure that the products were of high quality and with very low chemical residue value. Farmers had developed competencies in low chemical residue value tomatoes production. By combining the use of low levels of agrochemical inputs and natural methods of crop protection, farmers had produced tomatoes with less than 5% chemical residue values.

Participatory approach: FOs works in a participatory manner in its intervention approach. The idea to professionalize the production chain was identified in November 2006 during the follow up mission of SOS FAIM to NOWEFOR. After three days of field visits and discussions with leaders, the two parties saw the need to professionalize a limited number of agricultural speculations amongst which were market gardening, poultry, pig production, ginger, etc. These were chosen according to their economic contribution, the sustainability of the production systems and their contribution to organizational development. Farmer leaders, staff and SOS FAIM's partnership officer carried out joint reflection sessions to put in place the full project proposal and the budget. This process led to the definition of the methodology, the design of poultry houses and profitability thresholds for the production activities, the responsibilities of the farmers, the intervention zones and speculations and sustainability strategies amongst others. The farmers and their leaders contributed materially, financially and provided labour during execution of their self-help assistance projects. The local management committees and animators were to oversee the monthly review of project activities during monthly zonal meetings. The board of directors and executive committee steered the project execution, monitoring and evaluation in collaboration with SOS FAIM and staff.

Input capitalization fund: NOWEFOR had put in place a fund that should permit the farmers to continue to have access to productive resources after aid ends. To build this fund, farmers made financial contributions of up to 10 to 20% of the value of the inputs they received. Without contributions from the farmers, there is a risk of poor appropriation of the project. The approach also enhanced partnership and the development of a self-reliance spirit in the farmers. This fund had as objective to reduce the dependency on future assistance for the acquisition of inputs. An input capitalization bank account was created to host the fund. At the level of the community, the NOWEFOR savings and credit schemes assisted in the collection of the percentages. These percentages were centralized in the input fund's account. At the start of each farming season, community local animators assessed the collective needs of farmers and apply for funds to acquire the inputs as a revolving fund.

To build this fund further, a system of obligatory savings and commissions on group sales was set up and it served as the farmers' savings. All the commissions and savings were credited to the member's individual savings accounts. The commissions were placed in thetime savings account[1] and therefore can only be accessed at the start of farming seasons by the member. Increased savings permitted the farmer to get access to inputs, increase farm sizes, reinvest into the farming activity.

These findings agree with the Community Development Exchange (CDX, 2008) and Horton et al. (2004) who reported that technical skills would enable more people to play an active role in the decision making that affect their organizations. These findings tie with Penunia (2011:2-3) who highlighted that FOs are essential institutions for the empowerment, poverty alleviation and advancement of farmers and the rural poor as follows: Politically, farmers' organizations (FOs) strengthen the political power of farmers, by increasing the likelihood that their needs and opinions are heard by policy makers and the public. These findings confirm with those of Msuta and Urassa (2015:2343) and Shrestha (2015:3) who both highlighted that FOs help farmers gain skills, access inputs, form enterprises, process and market their products more effectively to generate higher incomes. By organizing, farmers can access information needed to produce, add value, market their commodities and develop effective linkages with input agencies such as financial service providers, as well as output markets. FOs can achieve economies of scale, thereby lowering costs and facilitating the processing and marketing of agricultural commodities for individual farmers. Marketing-oriented FOs can assist their members purchase inputs, equipment, meet quality standards and manage the drying, storage, grading, cleaning, processing, packing, branding, collection and transportation of produce.

Conclusively the contribution of FOs on the increased on improved input need assessment and supply skills, production skills and marketing skills was overall positive.

Contribution at the level of the organization

This section presents the contribution of FOs to group marketing and input supplies, evolution of technical staff and membership.

As revealed by Table 3, 27.14% of improved marketing skills and abilities of the beneficiary farmers increased compared to 11.42% of the non-beneficiary farmers. This could be explained by the fact that FOs aims at improving the living condition of farmers. This is usually done primarily by identifying agricultural speculations that are economically viable and facilitating the acquisition of technical and financial assistance to indulge in the

[1] This is a savings product in the NOWEFOR credit houses. Here the farmer saves a predetermined amount over a fixed period of time. He can only access the savings at the agreed period.

Figure 8. Reflection meetings with farmers on how to make the gardening subsector profitable.

production.

Experience of group marketing of tomatoes by market gardening farmers of Mforyah Integrated Farmers Union (MIFU) on the local mastery of the market and organized group marketing

Background

Farmers in Mforyah affiliated to NOWEFOR identified tomatoes cultivation as an important income generating crop in 2009. The farmers in Mforyah Integrated Farmers Union (MIFU) were organized into a gardening sector and received technical and financial assistance to get involved in the production of tomatoes and other assorted garden crops. The gardening sector mobilized a lot of youth farmers in the Mforyah community. It was therefore an important activity to increase the adhesion of youths to NOWEFOR. Tomatoes cultivation entails the used of agro chemical to combat pests. The farmers in this production subsector were trained to limit application of these chemicals to the minimum levels possible so as to chemical residues in the produce. There organic farming was the method being promoted in this subsector. Forty (40) young men and women were involved in the sector in 2009.

High supply and low prices for tomatoes

With the technical and financial assistance received from NOWEFOR, the farmers realized increased production. Each farmer moved from 6 buckets of 15 L of tomatoes per week to between 25 and 40 buckets of 15 L of tomatoes per week. The local markets in Mforyah as well as the nearby Bambui and Bamenda main markets were therefore flooded with tomatoes and this led to a price drop from 4000 F CFA per 15 L bucket to between 2000F CFA to 2500 F CFA. This situation was not comfortable as the farmers were not receiving satisfactory returns for their produce. The farmers in this production subsector reflected on how to make the production subsector more profitable as illustrated in Figure 8.

Two ideas came up namely how to reduce the supply of tomatoes in the local market and also how to pool the local farmers produce and look for external market outlets.

The farmers adopted a sequential production so as to limit the supply of tomatoes in the local market. Members of the market gardening subsector were grouped into five groups and planting calendar agreed upon to separate planting dates by weeks intervals among the subgroups. This meant that the farmers harvested at different intervals or times and in this way not all farmers took tomatoes to the local market at the same time.

In search for new market outlets, two market gardening sector members were sent to carry out market research in Douala and Limbe. Two bulk buyers were identified respectively in Douala and Limbe.

Pooling of tomatoes and group sales

The buyer in Douala showed a lot of interest and requested the farmers to send 225 kg of tomatoes by mid-December 2009 to be tested for chemical residues. The test on the tomatoes scored 91% while other producers who had also tendered to supply scored respectively 86 and 62%. The bulk buyer (leader price) agreed to buy the produce. The first command or order was placed in December 2009 for the supply of 2.5 tons of tomatoes at 3500FCFA per basket of 20 kg compared to the 2500FCFA at the local market. The contact farmer mobilized the other members of the production subsector and some of them pooled their tomatoes on the agreed date. This was collected and then delivered to the bulk buyer (Leader Price) in Douala.

Negotiating better marketing arrangements

The farmers experienced constraints in transporting the produce to Douala. These constraints were costs and handling of produce. The problem was presented to the buyer who agreed to take over the transportation aspect. Therefore, a new arrangement was arrived at which entailed farmers to mobilize their produce at the level of the village at the request of the buyer. The buyer then came to the village and paid for the produce and took them to Douala.

The farmers also observed that the perishable nature of the tomatoes meant that they should be the ones to determine when the tomatoes were available. This was also discussed and agreed upon with the buyer. New arrangements were made that the buyer gives the quantities needed for a period of six month and this was to be supply on a weekly basis. Therefore, a contractual agreement was signed between this farmer's organization and the buyer for a six months period on minimum quantities of tomatoes to be supplied monthly. The produce was supplied at a constant rate during the period.

Quality concerns

With the first supply of tomatoes to the buyer, 150 kg of tomatoes (about 10 buckets of 15 L) were rejected for poor quality. This prompted the group members to request for training on integrated pest management from NOWEFOR. Improved techniques on limiting agro-chemical applications to the minimum were dwelt upon and the farmers were fully capacitated in this area. The next supply of tomatoes scored 95.5% after the test. Also the supplies in March and April 2010 scored 100%. In April the farmers received a letter of congratulations from Equatorial Guinea and Gabon. The buyer also called on the farmers to maintain the quality with prospects of increasing the buying price in the future given the quality.

Supplies and incomes

Between December 2009 and September 2010, a total of 25.2 tons of tomatoes were supplied to this buyer. This brought a total income of 4,032,000FCFA to the market gardening farmers in Mforyah. The massive exportation reduced the abundance of tomatoes in the local market. Farmers producing other varieties for sell in the local market now experience better prices.

Impact of this group marketing of tomatoes in Mforyah zone and NOWEFOR

1. There was secured and regular income for farmers' tomatoes.
2. The farmers were able to acquire inputs as a group from Douala at low prices
3. Improved qualitative and quantitative production by group members (yields changed from 9tons/Ha to 14 tons/Ha)
4. The constant good quality of the tomatoes has prompted the buyer to request other produce namely water melon, sweet pepper, etc.
5. Most idled youth in the community have found employment in market gardening and increased their commitment (annual due contributions) in NOWEFOR activities.

Challenges and perspectives

The challenges and perspectives identified in this market gardening subsector were:

1. Sometimes the farmers are unable to supply the quantity demanded. There is therefore need to increase production while maintaining the quality.
2. This increased in production would there also enable the group contract other buyers to reduce the risk of relying on one buyer,
3. Other farmers from the organization have witnessed the regular income from gardening and have expressed the interest to join the gardening subsector. This means that there is need for assistance to train new farmers who become interested in tomatoes cultivation.
4. Resources are required to indulge in quality production of other produce being demanded by the buyer
5. The Mforyah group marketing experiences has been shared to other communities namely Nchum, Babungo, Batibo, etc. The experienced was first copied from Bambui Union of Sustainable Self-help Groups (BASSUG).
6. More 60 youths are now involved in market gardening production in Mforyah.
7. Production in the zone had increased and new prices of 4500FCFA per basket have been negotiated with buck buyers.
8. Annex business of packaging materials such as baskets is gradually increasing and creating jobs.

In a nutshell, this experience shows that pooling together farmers produce can enable them access special market segments. In this case organic tomatoes are seen to

Table 5. Distribution of respondents by membership strength.

Parameters and characteristics	Category of beneficiaries		
	Beneficiaries (%)	Non beneficiaries (%)	Total [280(100%)]
Membership strength			
Increased	128 (45.71%)	24 (8.57%)	152 (54.28%)
Constant	16 (5.71%)	76 (27.14%)	92 (32.85%)
Decreased	16 (5.71%)	20 (7.14%)	36 (12.85%)
Staff strength			
Increased	148 (52.85%)	8 (2.85%)	156 (55.71%)
Constant	16 (5.71%)	84 (30.00%)	100 (35.71%)
Decreased	8 (2.85%)	4 (1.42%)	12 (4.28%)

have insatiable demand compared to inorganic. This has also engaged a disadvantaged segment of the population (youths) in agriculture. The challenge faced by farmers now is how to increase production while maintaining the quality as they are a ready market. However, the practices involved in the process are not institutionalized. The farmers brought their tomatoes for group marketing at their free will. Also, the bulk buying of inputs at the level of the Douala was done by viable farmers who put money to pre-finance the purchased.

The contribution of FOs on the organization of group sales is presented in Table 4. From 2007 up to 2017 as illustrated in Table 4, the increase in pig, assorted gardening crops, and broilers production continued and the problem of marketing was posed. Once these produces were produced, it was bought at low prices by middlemen who in turn sell them in urban towns at a much better price. The challenge was how to increase the producer's own part of the income from these activities.

Based on this problem raised, NOWEFOR in bit to address this built the capacities of union leaders on improved marketing and group marketing techniques; put in place a marketing network and a marketing fund that would all facilitate the marketing process. After all the necessary measures were taken to ensure good marketing of farmers produce, the number of groups sales or marketing of pigs, assorted gardening crops and broilers as revealed by Table 4 increased from 40 in 2007 to 225 in 2010 making an overall increase of 82%. This increment could be explained by the fact there was easier access to information and trainings, harmonization of marketing strategies and the existence of marketing network that strived for better prices for farmers produce. As a result of this a large number of new producers joined the NOWEFOR unions, based in Based in Bafut, Mforyah, Nchum, Mundum 1 and Bambui. According to the Community Development Exchange (CDX, 2008) an empowered organization is one which is confident, inclusive, organized, co-operative and influential. It is therefore important to reflect on possibilities of committing members to supply during group marketing as well as participation in bulk buying of inputs.

These results also agrees with NOWEFOR (2017:2) who outlined that farmers organizations also play a vital role in providing training and sharing of experiences on production and marketing techniques; organization of wholesale purchases of inputs and group sales of farm produce; and the implementation of awareness practices that preserve the environment

This finding agrees Fongang and Forbah (2007:2) who reported that farmers' organization (FOs) in Bambui zone of the North West Region of Cameroon contributes in the facilitation of group marketing of tomatoes products for farmers. They further said between December 2005 and September 2006 a total of 25.2 tons of tomatoes were marketed as a group and this brought in a total income of 4,032,000 FRS to the farmers in areas.

It also agreeD with Gruère et al. (2009:39) who reported that farmer organizations tin South India play a critical role in the commercialization of underutilized plant species for the benefit of the poor and the conservation of agro-biodiversity.

Furthermore this finings agrees with Fischer and Qaim (2012: 1255), who reported that farmer organizations of small banana holder farmers in Kenya contribute as important catalysts for innovation adoption (trainings, improved varieties, etc) through promoting efficient information flows.

It stemmed from Table 5 that the 45.71% of beneficiary FOs membership rose/increased meanwhile only 8.57% of membership of non-beneficiary increased. The result reveals that 52.85% of the beneficiary FOs staff strength increased against 2.85% of non-beneficiary FOs. This implies that the aid from partners facilitated the increased in membership of FOs, number of trained leaders and the number of technical personnel of the federations.

The evolution of membership and technical staff of NOWEFOR are illustrated in Figure 9. The data in Figure 9 reveals that the membership and technical staff of NOWEFOR respectively rose from 923 and 2 in 2007 to 3100 members and 7 technical staff respectively in 2017.

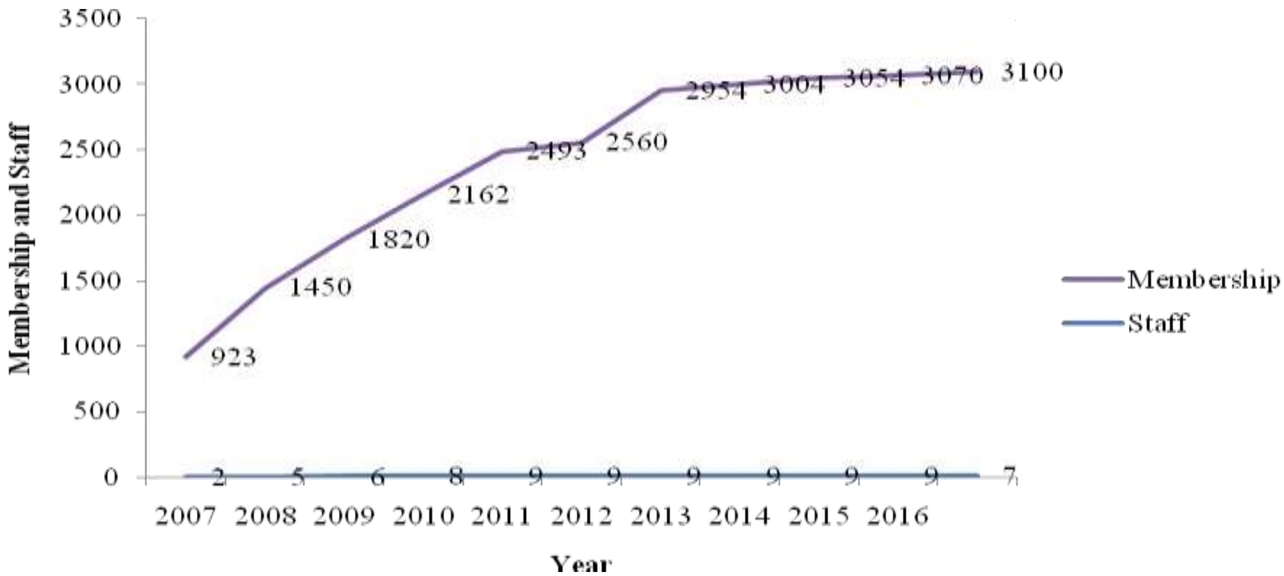

Figure 9. Evolution of membership and staff of NOWEFOR.

It could be concluded from Table 4 and 5 and Figure 9 that the contribution of FOs was overall positive on the increased in membership and technical personnel of the federation. These findings are in line with Czuba (1999) who reported that empowerment is a multi-dimensional social process that helps people gain control over their own lives. These results also affirms with those of Fongang and Fru mbangari (2017:139) who reported that FOs contribution to the promotion membership development and staff strength of organization in the rural community. Furthermore, these findings agree with those of Asante-Addo et al. (2016:1) who reported that farmer organizations in Ghana contributes or play an important role in training farmers in their activities and increasing membership in their organization.

These findings also agree with Arouna (2018:1) who report that FOs involved in group sales in Benin increased the farm income of rice farmers on average by USD 148/Ha and these increases in farm income were boosted by membership in a farmer group, training, and agreement on price. He also stressed that better training and well-functioning farmer groups facilitate group marketing.

Conclusion

Farmers' organizations play a vital role in reinforcement of the technical and organizational capacities of farmers. This study carried out from January 2018 to March 2019 in Mezam Division of the North West Region of Cameroon is therefore aimed at assessing the role of farmers' organizations (FOs) on the strengthening of the technical and organizational capacities of farmers and organization. Following the findings from the study, it can be concluded that beneficiary FOs members access more than is the case with non-beneficiary members as a result, this enable them to have improved skills in input supply (51.42%), production (38.57%) and market access (27.14%). Lastly, it is concluded that improved technical staff (52.85%), increased membership (45.71%) and improved group input supplies and group sales are strengthened in the organisation. This study concluded that farmers' organizations are very vital in the reinforcement of the technical and organisational capacities of farmers and organisations.

CONFLICT OF INTEREST

The authors have not declared any conflict of interests.

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REFERENCES

Asante-Addo C, Mockshell J, Siddig K, Zeller M (2016). Agricultural credit provision: What really determines farmers' participation and credit rationing. 5th International Conference of the African Association of Agricultural Economists, Addis Ababa, Ethiopia, September 23-26, P 25.

Benoit JB (2006). Evaluation of North West Farmers Organisation, 2nd evaluation, Unpublished, SOS Faim Luxembourg P. 15.

Bilal S (2014). Experimental and non experimental methods impact assessment. Innovation for agriculture. DME Impact evaluation workshop held on the 16th -20th at Kigali Rwanda jointly organized by the World Bank group, UK Aid, GAF SP and Department for International Development P 40.

Czuba CE (1999). Empowerment: What is it? An extension journal. Inc. ISSN 1077-5315, Haddam P 6.

Defang HF, Kana JR, Bime MJ, Ndebi G, Yemele F, Zoli PA, Manjeli Y, Tegui A, Tchoumboue J (2014). Socioeconomic and technical characteristics of pig farming in the urban and peri - urban zone of Dschang - West region of Cameroon. Discourse Journal of Agriculture and Food Sciences www.resjournals.org/JAFS ISSN: 2346-7002 2(1):11-20

Fischer E Qaim M (2012). Linking Smallholders to Markets: Determinants and Impacts of Farmer Collective Action in Kenya. World Development 40(6):1255-1268.

Fongang GHF, Fru Mbangari K (2017). Contributions of farmers' organizations to rural development: case of North West Farmers' Organization in Mezam Division, Cameroon, Journal of Agricultural Extension and Rural Development 9(7):129-142.

Fongang G, Forbah DN (2007). Valorisation d'expériences d'organisations paysannes « Accès au marché et commercialisation de produits agricoles ». - Forum régional Bamako, 16-18 janvier 2007 Inter-réseaux Développement rural – CTA, Retrieved from http://www.inter-reseaux.org/IMG on 08/10/2018, P 2.

Fotso KP, Meutchieye F, Andriamanalina SI, Youbissi A, Tchoumboue J, Pinta JY, Zango P (2014). "Caractéristiques socio-économiques et techniques de l'apiculture dans les Départements de Bamboutos, Mifi et Menoua, de l'Ouest Cameroun". Livestock Research for Rural Development P 6.

Gouët C, Leeuwis C, Van Paassen A (2009). Theoretical perspectives on the role and significance of rural producer organizations in development: implications for capacity development. Social and Economic Studies 58(3/4):75-109.

Gruère G, Nagarajan L, OliverKingc EDI (2009). The role of collective action in the marketing of underutilized plant species: Lessons from a case study on minor millets in South India. Food Policy 34(1):39-45

Hulme D (1997). Impact assessment methodologies for micro finance. A review, AIMS Brief No. 14:1-15. IMF, country Report, No. 03/249, P 35.

Koehler J, Zürcher C, Böhnke J (2007). Assessing the impact of development cooperation in North East Afghaistan: Interim report. Approaches and methods. Evluation, working paper, Ministry of external cooperation and development, Germany P 62.

Masud N, Yontcheva B (2005). Does foreign aid reduce poverty? Empirical Evidence from Nongovernmental and Bilateral aid. IMF Working Paper, WP/05/100, 31:31.

Msuta PB, Urassa JK (2015). The contribution of farmers organisations to smallholder farmers well-being: A case study of Kasulu district, Tanzania. DOI: 10.5897/AJAR2014.9261.

NOWEFOR Evaluation Report (2012). Evaluation of North West Farmers Organisation. SOS Faim Luxembourg, Bamenda P 25.

NOWEFOR Report (2017). Annual Activity of report. North West Region of Cameroon, SOS Faim Luxembourg,. Retrieved from https://www.biodynamics.com/files on 19/10/2018, P 30.

Republic of Cameroon (2015). Third General Census of Population and Housing 2005 - IPUMS Subset. CMR-2005-PHC-v01-M-v6.3-A-IPUMS, Central bureau of census and population studies, Yaoundé P 40.

Penunia ES (2011). The role of farmers' organizations in empowering and promoting the leadership of rural women. https://asianfarmers.org/the-role-of-farmers%E2%80%99-organizations-in-empowering-and-promoting-the-leadership-of-rural-women/

Shrestha RK (2015). Building social capital within the framework of agricultural at the School of Agriculture and Food Sciences. University of Queensland, Autralia P 314.

The Community Development Exchange (CDX) (2008). Teachers for Life: Empowering refugees to teach and share knowledge. The National Empowerment Network Guide P. 105.

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Full Length Research Paper

Evaluation of maize (*Zea mays* L.) performance under minimum and conventional tillage practice in two distinct agroecological zones of Uganda

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Maize is one of the major staple foods in Uganda, providing over 40% of Uganda's daily calorie consumption. Tillage practice is one of the crucial factors that influence crop productivity through maintenance of soil health. The aim of this study therefore is to validate the effect of tillage practice on the vegetative growth and yield of maize in Uganda. The trial was established on-station at Ngetta Zonal Agricultural Research and Development Institute, Lira district and on-farm in Njeru Town Council, Buikwe district. It was laid out in a factorial design with two tillage practices (conventional tillage, CT and minimum tillage, MT) as the main factors and two maize varieties (Longe 10H and Longe 5) as the levels. The results showed significant differences in plant height between the two tillage methods ($p < 0.001$). Conventional tillage practice had a higher mean maize grain yield per hectare compared to minimum tillage although the difference was insignificant ($p < 0.332$). Thus, the adoption of minimum tillage practice by farmers in Uganda would require investment in organic herbicides in order to control the weeds sustainably and boost maize productivity.

Key words: Conventional tillage, minimum tillage, vegetative growth, yield, maize.

INTRODUCTION

Among the most important cereals worldwide is maize (*Zea mays* L.) with the highest average yield per hectare (FAOSTAT, 2012) in developing countries, about 62% of the crop is consumed by humans as food, 34% as animal feed while the rest is as seed and industrial purposes (FAOSTAT, 2012). Due to its numerous uses, the crop is increasingly gaining an important position in the cropping system especially in Uganda where there is a rapid increase in the population (Karunatilake et al*.,* 2000). In Uganda, the North, Eastern and Central regions are the major producers of maize (Khurshid and Nawaz, 2006). However, a number of factors limit its production

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including poor farming practices (conventional and minimum tillage operations).

Tillage has been recognized as a factor that causes degradation of natural resources through soil erosion, contamination, decline in above and below ground biological diversity, deforestation, desertification, salinization, and greenhouse gas emissions (Le Guillou et al., 2019). Despite profound evidence of soil translocation processes, it was only in the 1990's that systematic research on this topic began (Van Oost et al., 2006). Conservation tillage, also known as minimum tillage is the use of agricultural practices that have minimal soil disturbances (Hossain et al., 2015). It is also defined as a farming system that employs a broad set of practices with a goal leaving some crop residue on the soil's surface to increase water infiltration and reduced erosion (Reicosky, 2015). Small resource poor farmers who have adopted conservation tillage methods cite the reduction in labor inputs and drudgery as major drivers for adoption (Andersson and D'Souza, 2014). Conservation tillage is also known to enhance soil fertility through reducing soil erosion and ultimately improving crop yields (Pittelkow et al., 2015).

Several types of conservation tillage such as minimum tillage, incomplete tillage, reduced tillage, and no tillage are practiced across the world. According to data gathered by the Conservation Technology Information Center (Anonymous, 2004), about 40.7% of total crop land on 45.44 million hectares was under conservation tillage system. Of that, no tillage was used on about 23.6% of land in the United States. Generally, reduced or no tillage provides minimum disturbance of the soil and leaves the surface covered with crop residues. The crop residues are not absolutely mixed and most or all of them remain on the top of the soil surface rather than being ploughed into the soil. They maintain a constant cover of organic material on the surface, which retains water and minimizes runoff, reduces erosion and sedimentation and improves water quality.

On the contrary, there is a common practice referred to as conventional tillage which deploys stirring up the deep layer of soil, incorporating plant debris and exposing the soil pests to sunshine for control, then lump breaking and levelling (Barbosa, 2015). It is also composed of harrowing which involves removing crop residues, subsoiling which breaks the compacted soil layer before levelling to form a fine seedbed.

In improving soil condition, tillage is a key factor and plays a significant role in improving maize growth and grain yield. A compacted soil layer, because of its high strength and low porosity, confines the crop roots in the top layer and reduces the volume of soil that can be explored by the plants for nutrients and water (Lipiec et al., 2003).

There is inadequate information on the effect of tillage methods on maize growth and yield in Uganda. This study was therefore to bridge the information gap in regards to the influence of tillage practices on the performance of maize in the country using Lira and Buikwe districts as case studies. The information generated will contribute to enhanced sustainable production of maize and thus guarantee both food and income security.

MATERIALS AND METHODS

Experimental sites

Two field trials were established in Uganda in the districts of Buikwe (Njeru) and Lira (Ngetta Zonal Agricultural Research and Development Institute). These sites represent two maize growing agro-ecological zones of Uganda, namely Lake Victoria Crescent and Northwestern Savanah Grassland respectively.

Lake Victoria Crescent is characterized by sandy clay alluvial soils with moist semi-deciduous forest, savannas, and swamps. The area receives rainfall ranging from 1750 to 2000 mm with bimodal rains comprising of April to May for the first ones and October to December for the second rains. Temperature ranges from 11 to 33°C. Climate is warm and wet with relatively high humidity and an average altitude of 1134 m above sea level. Northwestern Savanah Grassland is comprised of ferruginous sandy loam soils with intermediate savanna grassland and scattered trees. The rainfall received ranges averagely from 1340 – 1371 mm with bimodal rains followed by a dry spell for about 5 months. Temperature and altitude range from 15 to 25°C and 951 to 1341 m above sea level respectively (Gwandu et al., 2019). These two agro ecological zones were selected for the study based on their distinct ecological features or conditions and the history of maize growing.

Source and description of maize variety

Two maize varieties (Longe 5 and Longe 10H) were used for the study. These varieties were selected because they are the most widely grown in the two agro-ecologies. Longe 5 is a drought tolerant variety, sweet at green maturity, resistant to grey leaf spot and maize streak virus. It also has a good cob size and is suitable for low and mid altitude areas like Buikwe and Lira district. On the other hand, Longe 10H is high yielding, ideal for mid-altitude, bred with good drought and storage pest resistance. The seed was acquired from NASECO Seed Company in Uganda.

Experimental design and management

The experiment was arranged in a factorial design with 2 tillage practices (conventional tillage, CT and minimum tillage, MT) as the main factors and 2 maize varieties (Longe 10H and Longe 5) as the levels. It was replicated three times. Plot sizes were 5 m x 5 m with inter-plot spacing of 1 m. For no tillage, planting basins were dug after slashing at 0.6 m x 0.9 m spacing using a hand hoe and each basin was measuring 0.15 m (length) x 0.15 m (width) x 0.15 m (depth). Conventional tillage was done using a hand hoe. The first trial was planted in the second season (A) of September 2017 while the second one (B) was planted in the first season of March, 2018. It was done at 0.75 m x 0.25 m spacing with 4 maize seeds per hole for planting basins. Thinning was done to 2 plants each per hole for minimum tillage and conventional tillage.

Field data collection

Data was collected every fort night from two weeks after planting for

8 weeks on the following parameters; plant height, stem girth, number of leaves. Data on maize yield was later recorded at harvest.

Data collection procedure

10 plants were randomly selected from each of the plots. Each plant was observed and measurements done every after two weeks. Plant height, stem girth was measured using a string and the values read from a ruler. The maize cobs were harvested after 15 weeks and weighed on a weighing scale. The cobs from each of the plots were also threshed and weighed separately. The maize grains were washed clean and their moisture content (%) was obtained using a moisture meter (Infratec™ 1241 Grain Analyser - FOSS analytical). 100-seed weight (g) was also weighed per plot and the overall maize yield (tons/ha) was calculated (Kayode and Ademiluyi, 2004).

Data analysis

The data set for maize vegetative growth (plant height, stem girth and number of leaves) and maize yield were summarized and mean values obtained. ANOVA was done using GenStat version 12 statistical package to establish the effect of different tillage practices on maize.

RESULTS AND DISCUSSION

Effect of tillage practice on vegetative growth of maize

Most of the vegetative growth parameters did not significantly vary with location and season; therefore, the data was pooled together.

Plant height

Regardless of the tillage practice, the plant height increased with the age of the maize crop (Table 1). This could be attributed to the active cell division that occurs in the plant cells and thus cause a rapid change in size and length of the cells as the plants grow. This analogy is supported by research work of Nielsen (2000) who studied the growth and development of corn in Indiana, USA. He attributed the increase in plant height with age to the apical meristem which is an area of rapid cell division located at the tip of the corn stalk. Generally, plant height differed significantly with the tillage practice, variety and crop age ($p<.0.001$). For both varieties, minimum tillage registered higher mean height compared to conventional tillage (Table 1). This could be attributed to the ample soil cover for the minimum tillage plots which conserves soil moisture as well as the decomposition of the slashed residues that improve on the fertility of the soil and thus enhancing crop growth. These results are similar to those by Sornpoon and Jayasuriya (2013) who reported taller corn plants in the minimum tillage plots in Bangrakum district, Phitsanulok Province, Thailand. On the contrary, Kayode and Ademiluyi (2004) observed the least mean height in the minimum tillage plots in comparison with that in the tilled plots in Southwestern Nigeria. In fact, study by Drakopoulos et al. (2016) confirmed this trend when it registered significantly higher plant height of organic potatoes in the conventional plots as compared to the reduced tilled ones in Droevendaal, Wageningen, Netherlands. Also, Jokela and Nair (2016) reported no significant difference in plant height of organic bell pepper plants under minimum and conventional tillage plots in the Iowa region of United States of America.

Stem girth

Conventional tillage registered a much higher mean stem girth compared to the minimum tillage across the two varieties (Table 1). Mean stem girth did not vary significantly with tillage practice ($p<0.301$). However, generally, plants under minimum tillage had higher stem girth compared to those under conventional practice especially in Longe 5 (Table 1). This could be attributed to the presence of crop residues in the minimum tillage plots which decompose and form nutrients that result into better crop establishment. Similarly, a study by Jokela and Nair (2016) in the United States of America revealed that the stem diameter of the organic bell pepper plants under the minimum and conventional tillage plots was not significantly different. On the contrary, a study by Aikins and Afuakwa (2010) observed that the conventional tillage practices of disc ploughing alone, combination of ploughing and harrowing registered the biggest mean stem girth compared to the no tillage practice in cowpea rainfed fields of Kumasi region in Ghana.

Number of leaves per plant

Mean number of leaves per plant did not vary significantly with tillage practice ($p<0.075$) and variety ($p<0.910$). Higher mean number of leaves were recorded in minimum tillage plots compared to the conventional tillage plots in Longe 5 but the reverse was true for Longe 10H (Table 1). The number of leaves produced by a plant is directly proportional to the amount of photosynthate generated (Ridge, 1991). This literally means that this parameter was inconsistent and may not be used to explain the observed trend.

Effect of tillage practice on the yield components of maize

Mean grain yield (t/ha) and weight per 100 g of seed did not vary significantly with tillage practice. On the contrary, there was a significant difference observed between these two parameters with variety. A generally higher yield and weight per 100 g of seed was registered in conventional tillage plots compared to those of minimum

Table 1. Mean plant height/cm, stem girth/cm and number of leaves of two maize varieties under two contrasting tillage practices.

Parameter	Longe 10H		Longe 5H	
	CT	MT	CT	MT
Height (cm)	74.59[a]	82.90[b]	82.51[a]	83.27[a]
Stem girth (cm)	4.30[a]	4.28[a]	4.41[a]	5.29[a]
Number of leaves	9.65[a]	9.81[a]	9.63[a]	9.46[a]

Means denoted by a different letter indicate significant differences between treatments ($p < 0.05$).
Source: Research data analysis.

Table 2. Mean 100 maize seed weight (g) and mean maize yield (tons/ha) of two maize varieties under two contrasting tillage practices.

Season	Location	Mean weight per 100 g of maize seed				Mean maize yield (tons/ha)			
		Variety							
		Longe 10H		Longe 5		Longe 10H		Longe 5	
		Treatment							
		CT	MT	CT	MT	CT	MT	CT	MT
2017B	Buikwe	33.67[a]	30.67[a]	44.00[b]	43.33[b]	10.40[c]	9.00[c]	6.80[d]	6.00[d]
	Lira	34.77[a]	32.33[a]	43.70[b]	42.01[b]	8.60[c]	7.30[c]	4.90[d]	4.60[d]
2018A	Buikwe	37.67[a]	34.67[a]	45.67[b]	45.33[b]	9.10[c]	8.60[c]	7.10[d]	7.50[d]
	Lira	34.90[a]	34.33[a]	42.77[b]	43.93[b]	10.00[c]	8.70[c]	4.70[d]	5.80[d]

Means denoted by a different letter indicate significant differences between treatments ($p < 0.05$). Source: Research data analysis.

tillage (Table 2). This observed trend could be associated with increased soil loosening in the conventional tillage which enhances root penetration and nutrient uptake resulting into higher yields. This observation agrees with Salem et al. (2015) who reported an increment of 15.4% in the maize yield under the conventional tilled plots compared to the minimum tilled ones in the central region of Spain. Their study attributed this increase in yield to the tillage operations that improved soil physical properties like aeration and water retention. Rashidi and Keshavrzpour (2007) reported similar results after evaluating the effects of seven tillage practices on the yield components of maize under clay loam soil and they observed significantly higher grain yield in the tilled plots compared to the non-tilled ones. On the contrary, Armengot et al. (2015) observed no significant difference in the yields of organic sunflower and wheat between the reduced and conventional tillage plots in the arable lands of Switzerland.

Effect of tillage practice on the maize grain moisture content

There was no significant difference observed between the tillage practice and maize grain moisture content ($p<0.387$). However, the latter varied significantly with the location, variety and season ($p<0.001$). Generally, maize plants that were under conventional tillage recorded a higher grain moisture content compared to those that were under minimal tillage operations (Figure 1).

Abiotic factors like air flow, air temperature and air humidity which are largely influenced by the location and season significantly influence the grain moisture content (Hellevang, 2013). Grain moisture content can also be influenced by the inherent quality traits that are largely variety related factors.

Conclusion

This study revealed that conventional tillage practice resulted in higher maize grain yield compared to minimum tillage although the yield difference was insignificant. It however recommends research to be carried out on the influence of tillage practice on herbage yield in order to come up with a comprehensive package for the dairy farmers in Uganda. The study also guides that a deliberate research on the cost benefit analysis between the two tillage practices be carried out before any can be recommended to farmers.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

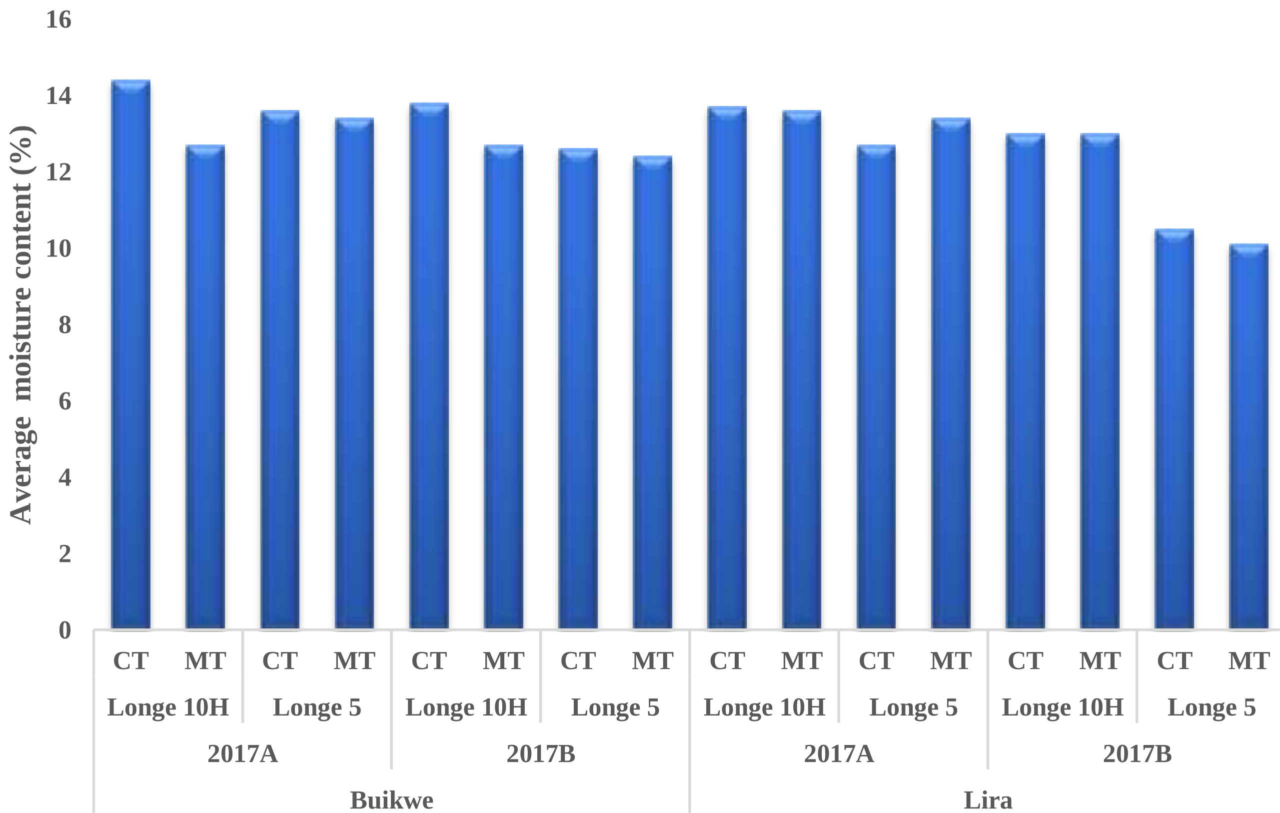

Figure 1. Mean percentage moisture content of maize grain under two contrasting tillage practices.
Source: Research Data (2017).

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REFERENCES

Aikins SHM, Afuakwa JJ (2010). Effect of four different tillage practices on cowpea performance. World Journal of Agricultural Sciences 6(6):644-651.

Andersson JA, D'Souza S (2014). From adoption claims to understanding farmers and contexts: A literature review of Conservation Agriculture (CA) adoption among smallholder farmers in southern Africa. Agriculture, Ecosystems and Environment 187:116-132.

Anonymous (2004). SAS Institute. SAS/STAT 9.1 User's Guide. SAS Institute, Cary, North Carolina, USA.

Armengot L, Berner A, Blanco-Moreno JM, Mäder P, Sans FX (2015). Long-term feasibility of reduced tillage in organic farming. Agronomy for Sustainable Development 35(1):339-346.

Barbosa M (2015). Sugarcane Agricultural Production, Bioenergy and Ethanol, Academic Press, pp. 35-51.

Drakopoulos D, Scholberg JM, Lantinga EA, Tittonell PA (2016). Influence of reduced tillage and fertilization regime on crop performance and nitrogen utilization of organic potato. Organic Agriculture 6(2):75-87.

FAOSTAT (2012). Food and agriculture data http://Faostat.fao.org (accessed October 6, 2015)

Gwandu C, Ochwo-Ssemakula M, Sseruwagi P (2019). Whitefly resistance in African cassava genotypes. African Crop Science Journal 27(2):213-228.

Hellevang KJ (2013). Grain drying. North Dakota state University Extension Service Book (701):231-255.

Hossain MI, Sarker MJU, Haque MA (2015). Status of conservation agriculture-based tillage technology for crop production in Bangladesh. Bangladesh Journal of Agricultural Research 40(2):235-248.

Jokela D, Nair A (2016). Effects of reduced tillage and fertilizer application method on plant growth, yield, and soil health in organic bell pepper production. Soil and Tillage Research 163:243-254.

Karunatilake U, Van Es HM, Schindelbeck RR (2000). Soil and maize response to plow and no-tillage after alfalfa-to-maize conversion on a clay loam soil in New York. Soil and Tillage Research 55(1-2):31-42.

Kayode J, Ademiluyi B (2004). Effect of tillage methods on weed control and maize performance in Southwestern Nigeria location. Journal of Sustainable Agriculture 23(3):39-45.

Khurshid K, Nawaz A (2006). Effect of tillage and mulch on soil physical properties and growth of maize. International Journal of Agriculture and Biology 8(5):593-596.

Le Guillou C, Chemidlin Prévost-Bouré N, Karimi B, Akkal-Corfini N, Dequiedt S, Nowak V, Terrat S, Menasseri-Aubry S, Viaud V, Maron PA, Ranjard L (2019). Tillage intensity and pasture in rotation effectively shape soil microbial communities at a landscape scale. Microbiology Open 8(4):e00676.

Lipiec J, Hatano R (2003). Quantification of compaction effects on soil physical properties and crop growth. Geoderma 116(1-2):107-136.

Nielsen LR (2000). Corn growth and development. What goes on from planting to harvest? Agry 97(07):1.1. https://www.agry.purdue.edu/ext/pubs/AGRY-97-07_v1-1.pdf

Pittelkow CM, Liang X, Linquist BA, van Groenigen KJ, Lee J, Lundy ME, van Gestel N, Six J, Venterea RT, van Kessel C (2015). Productivity limits and potentials of the principles of conservation

agriculture. Nature 517:365-368.
Rashidi M, Keshavrzpour F (2007). Effect of Different Tillage Methods on Grain Yield and Yield Components of Maize (*Zea mays* L.). International Journal of Agricultural 9(2):274-277.
Reicosky DC (2015). Conservation tillage is not conservation agriculture. Journal of Soil and Water Conservation 70(5):103A-108A.
Ridge I (1991). Plant Physiology. Hodder and Stoughon Educational Press. United Kingdom, P. 233.
Salem HM, Valero C, Muñoz MÁ, Rodríguez MG, Silva LL (2015). Short-term effects of four tillage practices on soil physical properties, soil water potential, and maize yield. Geoderma 237:60-70.
Sornpoon W, Jayasuriya HP (2013). Effect of different tillage and residue management practices on growth and yield of corn cultivation in Thailand. Agricultural Engineering International: CIGR Journal 15(3):86-94.
Van Oost K, Govers G, De Alba S, Quine TA (2006). Tillage erosion: a review of controlling factors and implications for soil quality. Progress in Physical Geography 30(4):443-466.

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Full Length Research Paper

Assessment and characterization of mung bean (*Vigna radiata)* bacterial brown spot in Eastern Amhara, Ethiopia

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Mung bean is one of the major early maturing pulse crop grown all over the world including Ethiopia. The production of the crop in Ethiopia, however, suffers from many diseases caused by bacteria. The study aims to assess the intensity and identify the major foliar bacterial and fungal pathogens of the crop. Purposively, 3 districts and randomly 90 mung bean fields were surveyed during the study period. Pathogenicity test, macroscopic and microscopic observations and biochemical tests were used for identification. Symptomatic of 33 diseased bacterial samples were initially isolated and purified on nutrient agar. Bacterial brown spot was found as important foliar devastating identified diseases, even if its distribution varied among localities. Water soaked, small, circular, brown lesions surrounded by yellow zones were observed in all bacterial brown spot isolates after 8 days of inoculation. Based on cultural and biochemical characteristics, bacterial isolates were identified as grams negative phytopathogenic bacteria called *Pseudomonas syringae pv. Syringae.* However, further characterization of both isolates and phenotypic characteristics of a large population of newly emerged *P. syringae* pv. Syringae from various host plants should capture the research attention. This is the first report on the occurrence of such disease in Ethiopia.

Key words: Bacterial brown spot, distribution, Eastern Amhara, identification.

INTRODUCTION

Mung bean is an annual food legume in the subgenus Ceratotropis in the genus *Vigna* (Lambrides and Godwin, 2007); it is the seed of *Phaseolus radiates* L., an annual herb of the Leguminosae family (EPP, 2004). It has green skin and is likewise called green bean. It is sweet in flavor and cold in nature (EPP, 2004). It is a standout amongst the most significant pulse crops, developed from the tropical to subtropical zones around the world (Khan et al., 2012; Kumari et al., 2012). It is a significant wide spreading, herbaceous and annual pulse crop developed

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generally by customary farmers (Ali et al., 2010).

Mung bean is originated from India and it has diversified to East, South, Southeast Asia (China) and some countries in Africa (Imrie and Lawn, 1991). It is a minor crop in Australia, China, Iran, Kenya, Korea, Malaysia, the Middle East, Peru, Taiwan and United States (Itefa, 2016). In Ethiopia, mung bean is a recent introduction and is found in different parts of the country (Teame et al., 2017). It is grown in the north eastern part of Amhara region (North Shewa, Oromia special *zone* and Southern Wollo), SNNPR (Gofa area) and pocket areas in Oromia region (Hararge) and Gambella (ECXA, 2014). Smallholder farmers in drier environments in Ethiopia grow mung bean. In North Eastern Amhara, farmers in some moisture stress areas have been producing mung bean to supplement their protein needs and to also effectively use scanty rainfall (Asrat et al., 2012). It is also grown in few areas of North Shewa and hence its consumption is not wide-spread like the other pulses. According to CSA (2017), Ethiopia produces about 42915.555 tonnes of mung bean annually and 1.136 t ha^{-1}. From the total annual production Amhara region produces 35297.25 tonnes annually. From the region, North Shewa and South Wollo are known to produce this crop. These two zones produce an average of 26277.54 tonnes annually. Generally, Ethiopia produces less when compared with other countries annual production like India 20.7 million tonnes and others (FAO, 2017). Among pulse crops, mung bean (*Vigna radiate*) is an important short-duration grain pulse crop with wide adaptability, low input requirement and ability to improve the soil by fixing atmospheric nitrogen (Sadeghipour, 2009). It is suited to a large number of cropping systems and constitutes an important source of protein in cereal-based diets (Khattak et al., 2001; Minh, 2014). Mung bean (*V. radiata*) is one of the utmost important edible food legumes of Asia. In India and some South Asian countries, the crop plays important dietary protein source in predominantly cereal rich diets. The crop serves as an alternative source of non-animal protein as was the case in some parts of East Africa during the outbreak of the Rift Valley Fever. Also, it is effectively cooked and does not cause flatulence (Pursglove, 2003). Mung bean has premium quality over other pulses due to its more palatable, highly nutritive, easily digestible and non-flatulent nature (Khan and Malik, 2001).

Recently, domestic consumption of mung bean has increased because of the rising popularity in Ethiopia, cultural foods and perceived health benefits due to high levels of certain minerals and vitamins (Tensay, 2015). In Ethiopia, this crop might be a promising source of human and animal food, especially during winter and summer seasons. It matures quickly and it does not compete with the main winter and summer season crops as wheat (*Trifolium alexandrenum*), sorghum and others. Farmers used mung bean in bordering areas to make the soil fertile without providing fertilizer on the land. So, farmers regard mung bean as traditional crop and cultivated by traditional farming ((Tensay, 2015). According to this author, farmers in Ethiopia used mung bean as food and fodder (36.8%), income generation, food and fodder (32%), food, fodder and improve soil fertility (22.4%), food, fodder, improve soil fertility and income generation (8.8%).

However, productivity of mung bean is decreased through biotic and abiotic stresses; including diseases, insect pests, drought stress, water stress, extreme high temperature, salinity stress as well as heavy metals (Waniale et al., 2012; Das et al., 2014). Ashrar et al. (2001) considered lower yield potential of mung bean is due to susceptibility to insect pests, diseases, undetermined excessive vegetative growth and small seed size. In Ethiopia, according to Tensay (2015) abiotic factors limiting yields of mung bean in terms of both quality and quantity are extreme drought, cold weather, untimely rain (rain after pod filling) and type of soil used for cultivating it. Biotic factors limiting mung bean productivity includes weeds, leave diseases, flying insects on pod and leave at any growth stage (Tensay, 2015). Chadha (2010) reported that all parts of crop plant including root, stem, branches, petiole, leaves, pods and seeds of the crops are vulnerable to disease and pest.

The most serious fungal diseases which infect the mung bean are root rot (*Macrophomina phaseolina)*, web blight, *Rhizoctonia solani* (*Thanatephorus cucumeris*), powdery mildew (*Erysiphe polygoni*), cercospora leaf spot (*Cercospora canescens*), anthracnose (*Colletotrichum lindemuthianum*) (Grewal, 1987). Serious viral diseases include the yellow mosaic viral disease (Karthikeyan et al., 2014) and leaf crinkle virus (Makkouk et al., 2003). It suffers from two major bacterial disease namely, bacterial leaf spot and halo blight (*Pseudomonas syringae* pv. *phaseolicola*) (Jitendra and Anila, 2018).

Among these bacterial brown spot caused by *Pseudomonas syringae* pv*. syringae* in dry bean is the major foliar disease to cause both qualitative and quantitative loss of mung bean (Kavyashree, 2014). It was reported as the most widespread bacterial disease of dry bean in South Africa and yield losses of up to 55% was noticed (Serfontein, 1994). The disease is seed-borne and mainly infects foliage and to a lesser extent, pods. Symptoms of brown spot initially appear as small circular, necrotic (brown) spots on the leaves, often surrounded by a narrow yellow halo (Howard et al., 2016). The reduction in photosynthetic activity and physiological changes are considerable, which leads to potential reduction in yield depending on the stage and time at which the diseases appears. However, the disease intensity depends upon the cultivar, growing period and environmental conditions. The average yield of the crop is limited due to different reasons in Ethiopia EPP (2004).

Understanding the association of disease intensity with cropping systems, crop combinations and management practices could help to identify the most important variables and to develop an integrated and sustainable management options (Zewde et al., 2007). Furthermore, isolation and diagnosis (verification) of causal agent are very important. However, despite the importance of the disease, there is no information available on the association, distribution and identification of the causal agents in Ethiopia. Particularly, Eastern Amhara National Regional state of South Wollo and North Shewa *zones* has received little attention from research, development efforts, inputs and no such extensive, quantitative survey and characterization has been done. Knowing such information is crucial to identify the diseases, to map the geographic distribution and determine the status of the disease in addition to providing baseline data to prioritize research problems. On the other hand, such information is of paramount importance as it can be related to yield loss and hence the economic impact of the disease (Ngugi et al., 2002). In addition, the disease rapidly spread and emerged in many mung bean planting areas and is causing a series of mung bean yield losses, making the disease a major threat to mung bean production in the areas. Thus, it is necessary to investigate the geographic distribution of foliar devastating bacterial and disease to know the intensity and characterize the causal agent of the disease. Therefore, the objectives of this study were to: (1) to assess the distribution of bacterial brown spot of mung bean, (2) isolate and characterize bacterial brown spot isolates of mung bean on the basis of cultural and biochemical characteristics and prove pathogenicity of some bacterial brown spot isolates of mung bean at green house.

MATERIALS AND METHODS

Description of the study areas

Field survey was conducted in field of mung bean growing areas during 2018 main growing season in two major mung bean producing zones, namely South Wollo and North Shewa administrative zones of Amhara National Regional state (Figure 1). The administrative district of North Shewa zone in which the survey carried out Kewot is located in the range of 10°41''-11°55' N latitude and 37°20'-39o57'35 '' E longitude at an altitude of 1120 - 1380 m above sea level (m.a.s.l). The area has an average annual rainfall of 1007 mm, with short rain between March and April and main rainy season between June and September, and annual mean minimum and maximum temperatures of 16.5 and 31°C, respectively (BoA, 2000).

The administrative districts of South Wollo zone for the study area were Kalu and Tehuledere. Kalu is located in the range of 10°55'20''-12°32'6 '' N latitude and 39°45''43'-39°56''27'E longitude. The climate of Kalu varies from dry sub-humid to semi-arid. The annual average rainfall of the district ranges from 750 to 900 mm. The annual temperature also ranges between 25-35°C. The altitude of the district ranges from 1400 to 2467 m above sea level (KDOA, 2010). Tehuledere district characterized by diverse topography is located in the range of 11°18'21''-11°22'19'' N latitude and 39°38''51'-39o44''2'E longitude. The annual temperature varies from 15 to 20°C. The average annual rainfall is 1030 mm. The highest elevated spot of the district reaches 2,928 m.a.s.l. The lowest elevated point has an altitude as high as 1,400 m.a.s.l.

Disease survey and sample collection

A total of 90 fields were surveyed and 15 fields per district were assessed. 33 bacterial samples were considered for the total fields. Incidence and severity were recorded based on the appropriate scales for each disease of each field. The survey was done at two growth stages (vegetative and flowering) of the crop. Geographic features like latitude, longitude and altitude were recorded from all surveyed areas using handheld Global Positioning System (GPS), to trace back the specific locations and symptoms of each disease. Additionally, parameters such as soil condition, cropping system, adjacent crop, previous crop, altitude and growth stage were recorded to determine the relationship with disease intensity. Districts were selected purposively based on intensity of bean production and disease problems, while fields were randomly selected at intervals of about 5-10 km along the main and accessible road sides. Fields were assessed according to Sseruwagi et al. (2004), by walking along in two diagonals (X-Fashion) of mung bean fields at spots of 0.5 m × 0.5 m (0.25 m^2) quadrat. Leaf and stem samples were collected as a sampling unit. Disease prevalence was determined by ratio of number of locations showing mung bean diseases to total number of locations/fields assessed for individual diseases and expressed in percentage. Field incidence of each field for each was calculated by totaling the number of plants with symptoms and converting to percent. Diseases severity was rated from 25 selected plants per field, by scoring five representative plants for each five spaced quadrats using standard scales for each disease.

Sample collection was performed immediately after the appearance of the initial symptoms in early August and September in 2018 main growing season. Samples showing bacterial and fungal symptoms during the survey were collected and labeled well in perforated polyethylene bags. Thirty three and twenty five samples showing bacterial and fungal symptoms were taken and transported to Ambo Plant Protection Research Center and the specimens were maintained in refrigerator at 4°C until isolation was carried out.

Disease severity rating

Disease severity rating was evaluated on each plant using a modified 1 to 9 International Center for Tropical Agriculture scale, where 1 = 0% foliage affected, 3=2% foliage affected, 5 = 5% foliage affected, 7 = 10% foliage affected and 9 = 25% foliage affected (Van and Pastor-Corrales, 1987). Further, these scales were converted into Percent Disease Index (PDI) by using formula given by Wheeler (1969).

$$\text{PDI} = \frac{\text{Sum of individual rating}}{\text{No. of leaves assessed} \times \text{Maximum disease grade}} \times 100$$

Isolation of bacterial brown spot

Isolation, pathogenicity test and biochemical characterization of BBS isolates were conducted at Ambo Plant Protection Research Center (APPRC). Thirty three samples showing bacterial symptoms taken were grown on the plating media. Nutrient agar (NA) and nutrient broth (NB) were used as plating media (Lelliott and Stead, 1987). Leaf and stem samples were washed with running water thoroughly and approximately, 4 × 7 mm sized small tissue segments cut from active tissue margins of lesions. Small symptomatic pieces of samples taken from leaves and stems were surface sterilized by 1% sodium hypochlorite solution (laundry bleach) for one minute, 70% alcohol for one minute and rinsed in sterile distilled water three times of one minute each (Geiser et al., 2005; Summerell et al., 2006) and the prepared samples were macerated in sterile distilled water and crashed using laboratory mortar and pestle. Finally, the filtrates were diluted using sterile distilled water and wait for 30 min and allowed the bacteria to float and release from the tissues and then each suspension of the plates were streaked with sterile wire loop on the agar media. The plates were incubated at 30°C for 24 to 48 h; while each plate was examined for culture growth. Those mixed bacterial colonies were purified and grown on nutrient agar. Finally, eighteen bacterial isolates based on similar identity showed on growth media were preserved and used for greenhouse and laboratory analysis. Preserved isolates were stored in 20% glycerol at -80°C.

Identification of bacterial brown spot

Pathogenicity test

Determination of pathogenicity and fulfillment of Koch's postulate is a very important step in the identification of phytopathogenic bacteria. Pathogenicity testing of the obtained isolates were checked by artificial inoculation of leaves of mung bean using the methods described by Lelliott and Stead (1987) and Klement (1990). The test was performed on two week old seedlings of N26, which appears highly susceptible to the pathogen in the affected field. Seeds of N26 variety were provided by Sirinka Agriculture Research Center. Seeds were sown in pots filled with a 3:1:1 mixture of clay soil, sand and FYM respectively as adopted by Suli et al. (2017). Five seeds per pot were sown. The planted pots were placed in the greenhouse with a temperature of 23 to 30°C. For preparing inoculum the representative isolates were cultured on nutrient agar (NA) medium for 24 h. Suli et al. (2017). The inoculum was made by suspending the bacterial cells in sterile tap water to an approximate cell concentration of 1×10^8 CFU/ml measured using McFarland standards at wavelength of 600 nm by ultra-spectrophotometer by the methods stated in Žarko et al. (2017). Inoculation was done with injection of hypodermic needle on the lower surface of leaves and the points of inoculation were sealed with parafilm to prevent entry of external contaminants as reported by Firdous et al. (2009). Similarly control plants were sprayed with sterile distilled water for comparison. After inoculation of plants the relative humidity was maintained by covering the pots with polyethylene bags, watering of the bottom of the pots and sacks placed on the floor of greenhouse. Pots with polyethylene were kept for 96 h. for the bacteria to enter into the tissues of plants. Re-isolation was made after eight days of inoculation and comparison was made between the re-isolated culture and original culture for better confirmation (Jamadar, 1988).

Morphological characterization of bacterial brow spot

As colony morphology on agar surface aids to identify the bacterial isolates, each isolate from colonies of characteristic shape, size and appearance were observed. Characteristic features of the isolate organism were observed by macroscopic (on nutrient agar) and microscopic observations according to Goszczynska et al. (2000) and Aneja (2003). A loopful of culture from overnight grown was streaked on the surface of nutrient agar and was incubated at 30°C for 24 h. Colony morphology, color, texture, margin (consistency) were observed. Shape of the isolate was identified by making simple staining method followed by its observation under light microscope. Bacterial smear was stained with methylene blue dye and examined. Microbial cells were observed for their shapes like rod or cocci or spiral. Gram staining was performed to look for the gram's nature of the isolates. Purple coloured cells remain grams crystal violet and were called gram positive bacterium. Pink coloured cells lost primary stain and picked up safranin colour and were called as gram negative bacterium (Schaad, 1980).

Biochemical characterization of bacterial brow spot

Nutrient agar (NA) and nutrient broth (NB) were used as plating media (Lelliott and Stead, 1987). Eighteen bacterial brown spot isolates were selected for biochemical tests based on similar morphological and growth characteristics on selective media. These biochemical tests were done three times for better confirmation.

KOH solubility test

KOH solubility test was performed by the method of Fahy and Hayward (1983) using 24 to 48 h old culture. Two drops of 3% KOH placed on to glass slide and the colonies of test pathogen were stired into the solution clean loop for 5 to 10 seconds. When the solution was viscous enough to stick to the loop, causing a thin strand of thread like slime stretched up, the result recorded as positive. The results were recorded and used for identification of isolates.

Oxidase test

Filter-paper saturated with 1% Kovac's oxidase reagent, (tetra-methyl-p-phenylenediamine dihydrochloride) and placed in a clean Petri dish to look for cytochrome enzymes. A suspected colony of bacteria from NA transferred with wooden stick to the filter paper and rubbed onto the reagent for 30 s. Isolates developed blue or deep purple colours within 30 s were considered as positive for Cytochrome oxidase (York et al., 2004).The results were recorded and used for identification of isolates.

Catalase test

Catalase test was performed by adding 1ml of a 3% solution of hydrogen peroxide to glass slide, a loop of fresh culture grow on NA medium were added into the solution by the method described by Sands (1999). Release of bubble from the culture was recorded as catalase positive. The results were recorded and used for identification of isolates.

Levan production

To test for the levan production of the isolates, a nutrient agar plates containing 5% sucrose were streaked by the test isolates and incubated for 3-5 days at 30°C. until heavy growth occur. Levan produced when colonies were convex, white, domed and mucoid (Fahy and Hayward, 1983). The results were recorded and used for identification of isolates.

Nitrate reduction

Based on the Dickey and Kelman (1988), the ability of the isolates to reduce nitrate to nitrite was evaluated in a test medium that contains NO_3, 1 g; peptone, 5 g; yeast extract, 3 g and agar, 3 g in 1 L distilled water, sterilized at 120°C for 15 min in tubes. Each isolate were inoculated by stabbing and sealing with 3 ml sterilized molten agar to avoid false positives and were incubated at 28°C. The growth of each bacterial isolate and bubble formation beneath the upper agar layer was observed and recorded as positive result for nitrate reduction three, five and seven days after inoculation.

Aesculine hydrolysis

Aesculine medium containing plates was streaked by the test isolates and incubated at 20°C. for 2-5 days. Dark colour developed indicates the presence of β-glycosidase activity then, recorded as positive and negative no dark colour developed. The results were recorded and used for identification of isolates (Goszczynska et al., 2000).

Hydrogen sulfide (H_2S) production

H_2S production was detected according to Aneja (1996) by using sulphide indole motility (SIM) agar medium (peptone, 30 g; beef extract, 3 g; ferrous ammonium sulfate, 0.2 g; sodium thiosulphate, 0.025 g and agar, 3 g in 1 liter distilled water autoclaved at 121°C. for 15 min). The isolates were inoculated by stabbing and by incubating at 28°C. for 48-72 h. The presence of black coloration along the line of stab inoculation were recorded as positive for H_2S production.

Gelatin liquefaction

This test was performed according to methods described by Dickey and Kelman (1988) by employing gelatin medium that contains beef extract, 3 g; peptone, 5g and gelatin, 120 g in 1 L distilled water, poured into test tubes and autoclaved at 121°C. for 15 min and cooled without slanting. The media was stab-inoculated with each isolate grown on YPSA medium for 48 h and that was incubated at 28°C. Three and seven days after incubation, each isolate were evaluated for gelatin liquefaction. The isolates in test tubes were kept at 4°C. for 30 min and gently tipped immediately. A medium that flows readily as the tube is gently tipped was considered as positive for gelatin liquefaction.

Starch hydrolysis

The isolates were streaked on starch agar medium (starch soluble, 20 g; peptone, 5 g; beef extract, 3 g; agar, 15 g in 1 L distilled water with pH 7 and autoclaved at 121°C. for 15 min) to evaluate their ability to hydrolyze starch (amylase production). The plates were incubated at 30°C. for 6 days and starch hydrolysis was observed by flooding the plates with Gram's iodine solution for 30 s. The appearance of clear zone around the line of growth of each isolate indicated starch hydrolysis (Aneja, 1996).

Potato rot test

Fresh potato tubers were washed, alcohol flamed, peeled and sliced into approximately 7 mm width. The slices were placed in 90 mm diameter petri-dishes and sterile distilled water were added to a depth of half the slice. One hundred micro-liters of a 24 h. old bacterial suspension in nutrient broth was pipetted into a 3 mm diameter well on the center of each slice. Positive results was indicated by decaying of potato beyond the point of inoculation; while lack of rotting suggested negative results. Negative control and un-inoculated nutrient broth was used as positive control (Ignjatov et al., 2007).

Tween 80 hydrolysis

Fatty acid esterase activity was tested by streaking the bacterial cell mass onto a fresh nutrient agar medium containing calcium chloride and Tween 80, a polymer consisting of polyoxy-ethylene-sorbitanmonooleate (Sands, 1990). The medium was prepared from peptone, 10 g; $CaCl_2$ dihydrochloride, 0.1 g; NaCl, 5 g; agar, 15 g; and distilled water, 1 L; with the pH adjusted to 7.4. Tween 80 was autoclaved separately and added with 10 ml/L and mixed before plating. Incubation was made at 28°C. for up to 7 days (Fahy and Hayward, 1983). An opaque zone of crystals around a colony was recorded as positive reaction for hydrolysis of Tween 80.

Fluorescent and non-fluorescent test

Finally, the above-mentioned Levan, Aesculine, and Gelatine liquefaction were used to differentiate the test isolates into fluorescent and non-fluorescent *Pseudomonas*. The biochemical tests positive reaction were to indicate the fluorescent *Pseudomonas* isolates. Additionally, the fluorescent Pseudomonas produces a yellow-green to blue fluorescent pigments on iron-deficient media (KB media).The results were recorded and used for identification of isolates (Goszczynska et al., 2000).

Carbohydrate utilization

The utilization of Carbohydrates as a sole source of carbon energy is useful in the identification of bacteria particularly *pseudomonads*. Carbohydrate utilization was tested for by using a basal medium and the results were recorded daily for up to 8 days (Hildebrand, 1998). Stab inoculation of bacterial suspension on basal medium containing mannitol, sorbitol, inositol, dulcitol, lactose, sucrose and maltose were tested for their result. Carbon sources were used to distinguish Pss from *P. syringae* pv. *Phaseolicola* isolates. The pH was adjusted to 7.2 with 40% of NaOH. Yellow colour was recorded as positive result.

Data collection and analysis

Data collected from the survey was coded and checked for consistence and completeness and analyzed using SPSS statistical procedure. Descriptive statistics were used to summarize the data. Analysis was conducted by disaggregating important relevant information by districts and *zones* so that comparison could be made. Pearson correlation coefficient was used to know the association of diseases epidemics and biophysical factors like soil condition, adjacent crop, previous crop and cropping system.

RESULTS AND DISCUSSION

Assessment of bacterial brown spot of mung bean

The results of the field survey conducted at South Wollo and North Shewa *zones* and laboratory works done at Ambo Plant Protection Research Center are presented in here under. A baseline field survey prevailed that, among the different foliar diseases of mung bean bacterial brown spot caused by *P. syringae* pv. *syringae* was prevalent.

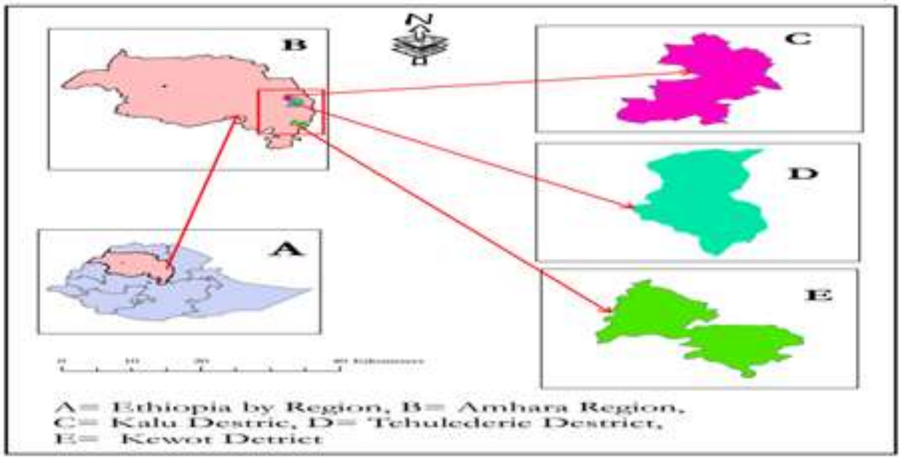

Figure 1. Map of the study areas (Kalu, Tehuledere and Kewot Districts).

Difference in intensity of mung bean diseases in the surveyed belt areas in Eastern Ethiopia was observed. There was variation with respect to disease incidence and severity across the surveyed areas. The variation of diseases intensity in various localities might be attributed to the climatic factors like temperature, relative humidity and distribution and amount of rain fall followed by cultural practices like sanitation and other suitable management practices. Scheuermann et al. (2012) also reported the same result.

The diseases was prevalent and widely distributed in all mung bean growing fields studied regardless of altitude, cropping system, soil condition, previous and adjacent crop. The distribution of this destructive disease in all surveyed areas might be either due to the environmental conditions which promote developments of the disease and/or due to presence of diversified causative pathogen across different mung bean growing areas. Variation in disease prevalence and intensity across locations would be attributed to prevailing environmental conditions and crop management practices, which was reported by Scheuermann et al. (2012).

Likewise, the high disease epidemics saw in all areas may be related to the poor cultural practices adopted by smallholder producers in the areas including utilization of low quality farmer-saved seed sources, absence of harvest turn and poor management practices exacerbated by helpful environmental conditions for disease development. High mean disease intensity observed in all agro-ecologies might be associated to the poor cultural practices adopted by smallholder farmers in the area including use of poor quality farmer-saved seed sources, lack of crop rotation and poor management practices exacerbated by conducive environment conditions for disease development (Njingulula, 2014; Kijana et al., 2017). Dependency on own seed sources could result in the build-up of inoculum and significantly contribute to the development of disease epidemics (Wachenje, 2002; Mwangombe et al., 2007). The amount of inoculum present in each farm, level of field sanitation and type of cropping system employed might also have influence on the overall disease distribution and epidemics (Stenglein et al., 2003; Mwangombe et al., 2007).

The findings of the study revealed that disease intensity of bacterial brown spot was more pronounced at altitudes greater than 1660 m.a.s.l. This might be partly due to the high rainfall and relative humidity common in the areas, which could favor diseases infection and epidemic development. Muedi et al. (2015) also obtained high disease incidence and severity of bacterial brown spot in altitude ranges of 1350-1735 m.a.s.l. due to high rainfall and relative humidity. The disease was noticed both at vegetative and flowering stages. At stage of vegetative mean incidence of the disease was observed as 45, 39 and 35% at Kewot, Kalu and Tehuledere districts respectively (Figure 2). The mean severities were 15, 12 and 10% from districts of Kewot, Kalu and Tehuledere respectively (Figure 2). The mean prevalence of bacterial brown spot at vegetative stage was recorded as 80, 66.6 and 66.6% at Kewot, Kalu and Tehuledere respectively (Figure 3). At flowering stage, the survey revealed that the bacterial brown spot disease was severe in Kewot and Kalu districts. The mean disease incidence of bacterial brown spot on mung bean showed 80.6, 83.3 and 74% at Kewot, Kalu, and Tehuledere districts respectively at flowering stage (Figure 2). Among districts surveyed, mean maximum severity of bacterial brown spot of mung bean was noticed at Kalu (50.73%) followed by Kewot (42.07%) (Figure 2). The mean least severity of the disease was recorded from Tehuledere district (36.67%) (Figure 2). The mean prevalence of bacterial brown spot was 100 % in all districts surveyed at flowering stage (Figure 3). This implies that the growth stage has a paramount importance for the distribution of the disease.

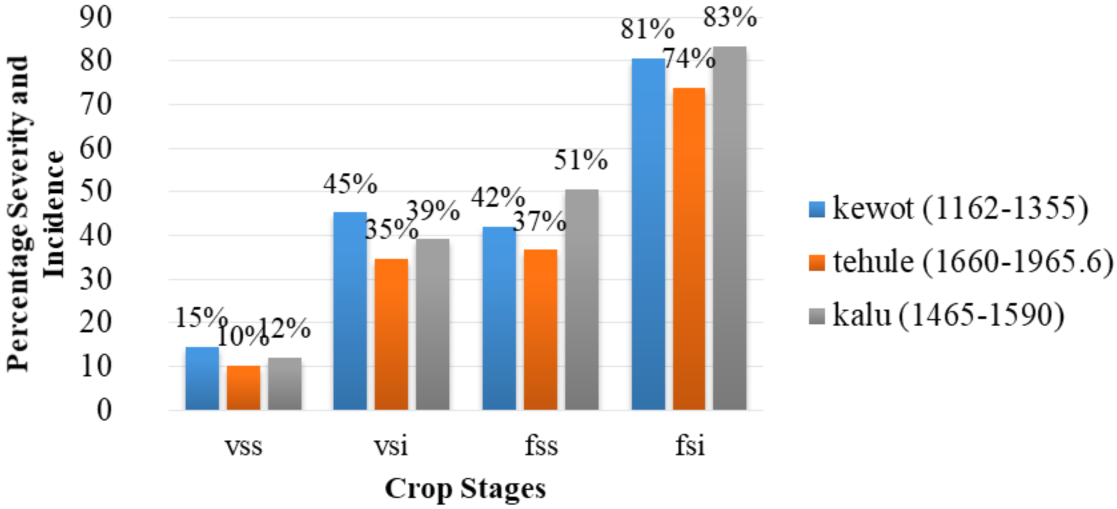

Figure 2. Bacterial brown spot diseases distribution with growth stages in the study districts with respect to altitudinal ranges.

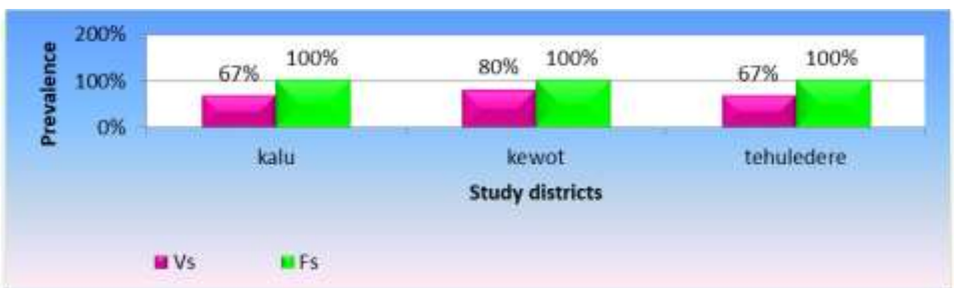

Figure 3. Prevalence of bacterial brown spot in the study areas at vegetative and flowering stages. Vs = Vegetative stage; Fs = Flowering stage.

Some of the mung bean growers were used chemical spray as diseases management option, but some growers were found to apply cultural practices like rouging and cutting of diseased plants and plant parts as disease management schemes to reduce rapid pathogen dispersal among plant canopies. Most of the farmers do not use pesticides as well as other protective mechanisms to prevent the negative impacts of pests and diseases associated with producing the mung bean. About 97% of the farmers were found not using pesticides for the management of bacterial brown spot at vegetative stage and 45% of growers were found using pesticides for the management of diseases with having no awareness on the type of pesticides to be applied (Table 1). Eighty eight percent of the grower's field were weed infested and 11% of fields were weed free (Table 1). With regard to cropping system, 88% of inspected farms were sole cropped and the remaining 12 % was intercropped with maize and sorghum (Table 1).

Regarding to previous crop, 13, 28 and 57% of fields were observed to have fallow, pulses and cereals respectively (Table 1). About (64%) of the adjacent crops observed in accordance with mung bean fields were cereals (Table 1). The rests (15%) and (20%) of fields were pulses and fields with no crops (Table 1). Some other mung bean disease and insect pest were also observed in association with the above diseases. Yellow mosaic virus and foliage beetle (*Ootheca* spp.) of mung bean were observed in all districts.

Association between biophysical factors and disease parameters

The correlation coefficient (r) between bacterial brown spot severity, incidence and variables like planting stage (vegetative-flowering), previous crop (fallow-cereal-pulse), soil condition (wet to dry), altitude (from low to high), adjacent crop (pulse- cereal-none) and cropping system (mono-intercropping) showed a relationship at

Table 1. Number of fields observed with different cultural practices.

Cultural practices	Number of fields observed in %
Pesticide used at vegetative stage	97.8
Inter-cropped fields	22.2
Sole-cropped fields	12.2
Fallow lands	13.3
Previous cereals	57.9
Previous pulses	28.8
Weed infested fields	88.8
Adjacent fields with pulses	15.5
Adjacent fields with no crops	20.01
Pesticide used at flowering stage	45
Adjacent fields with cereals	64.4
Fields with not weed infested	11

vegetative and flowering stages. Except growth stage and previous crop at flowering stage as well as previous crop at vegetative stage, the other variables association were negative with disease parameters at both stages (Table 4 and 5). Growth stage and previous crop showed positive association with disease severity and incidence at flowering stage (Table 4) and also previous crop showed a strong positive association with disease intensity at vegetative stage (Table 5).

Positive association between bacterial brown spot epidemics and growth stage has been observed at flowering stage (Table 4). This indicates that growth stage might have an influence to the disease epidemics. This might be because of the expansion in leaf canopy, due to the important relative humidity for the advancement of bacterial brown spot. Similarly, Robert (2009) directed the advancement of bacterial brown spot during the mid-vegetative to early flowering stage is because of the relative moistness due to expanded covering.

Positive association of previous crop at both stages with disease severity might be due to the pathogen ability to have a resident phase on both host and bean crop residue that may be an overwintering source for the pathogen (Tables 4 and 5). The epiphytic nature of the pathogen on weeds could result to grow and survive on the main host. Pathogen with a large host range has an increased chance of survival so that some plant pathogens may survive in alternate hosts without causing disease until they come in contact with the main host. Robert (2009) also reported the same finding with this result.

Negative connection of cropping system with disease severity for both stages might be attributed from the increase in spatial distance between host plants, which might inhibit free dispersal of pathogen and suppress weeds responsible for the build-up of high humidity under the canopy (Tables 4 and 5). It might also be due to rotating crops with non-host crop prevents the buildup of large population of pathogens. The factors result from the system itself and include change in microclimate, reduced host density, induced resistance and competition. Each of the factors may have a minor role in affecting disease. In the system, because of shading by the associate crop, temperature is relatively lower which is known to delay development of BBS. Thus, the bacterial multiplication and movement within the plant cells could be reduced and lower the inoculum. This finding is in line with Muedi et al*.* (2015) who stated that bacterial brown spot is prevalent where dry bean mono-cropping was practiced. There are also several studies with regard to the inhibition potential of intercrops against different pathogens in various pathosystems (Chemeda, 2003; Altieri et al., 2005; Habtamu et al., 2015).

The association of disease severity and adjacent crop showed negative and significant association at flowering and vegetative stage respectively at significance level (Tables 4 and 5). It might be due to the unavailability of diseased plants which provide the inoculum responsible for disease outbreaks in nearby bean fields. The absence of plants close to other fields that were infected in the past could be partly attributed for low disease epidemics. According to Ocimati et al*.* (2018), alternate and volunteer hosts in the neighboring fields are the source of inoculums for bacterial brown spot outbreaks.

Although altitude showed negative relationship with intensities of disease at both growth stages, it was not significant (Tables 4 and 5). It might be due to the suitable environmental conditions favoring BBS development and/or there might be variable pathogen races in the surveyed areas, which enable the pathogen to widely infect the host regardless of elevation differences. It could be also due to ideal moderate to warm temperature as favor for epidemics and severe localized disease outbreaks of bacterial brown spot. If environmental conditions are known to favor disease

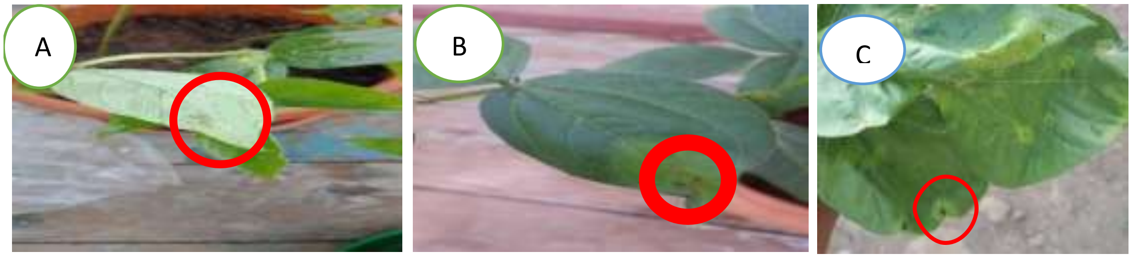

Figure 4. Water soaked on the underside of the leaves, (B&C): small, circular, brown lesions surrounded by yellow zones at green house and field condition.

development, there might be high disease incidence and severity even in high altitude areas ranging 1625-2000 (Muedi et al., 2015). Schwartz et al. (2019) also reported the incidence and severity of bacterial diseases such as halo blight and bacterial brown spot outbreaks are most serious when temperatures are moderately cool and humidity is high.

Soil condition relation with disease severity directed negative connection at flowering stage and vegetative stage (Table 4 and 5) that might be due the presence of dry soil condition during the assessment time for the spread of bacterial brown spot of mung bean. Indirectly wet soil is good for the spread of bacterial brown spot of mung bean. The same result has been reported by Muedi et al*.* (2015) who stated that avoiding working when soil is wet will decrease the disease severity and development as well as working when the soil is dry, which could result in reduction of disease epidemics.

Symptomatology of bacterial brown spot

Based on this study macro and microscopic observations, pathogenicity and biochemical tests revealed the pathogen called as *P. syringae* pv. *Syringae (*Pss) that leads the disease called bacterial brown spot of mung bean, which is newly emerging disease of the crop. *P. syringae* pv. *syringae (*Pss) has a wide host range groups even in different genera, infecting a number of host plant species. The typical leaf spot symptoms by all isolates were observed on leaves after eight days of inoculation. The symptoms of the leaf spot disease were characterized by production of small circular brown lesions surrounded by yellow zones (Figure 4B and C) and evidence of water soaking was visible on the underside of the leaves (Figure 4A). Linear necrotic lesions among veins were formed as a result of coalescing of lesions. On the affected leaves old lesion center fall out leaving shot holes was observed.

Bacterial brown spot is the most economically important diseases of the processing beans and woody plants Goszczynska et al. (2000). It was commonly reported on dry bean and stone fruit trees as *P. syringae* pv. *Syringae* (Pss)*.* Brown spot of bean (*Phaseolus vulgaris* L.) (Goszczynska et al., 2000; Serfontein, 1994), bacterial canker of stone fruit trees, bacterial blossom blight or blast of pear and citrus blast and black pith (Goszczynska et al., 2000) were noticed. Saettler (2005) reported the disease has wide host ranges, infecting more than 180 host plant species including woody plants and weeds.

In line with this Howard et al. (2016) showed initial symptoms of brown spot first appear as minor rounded, necrotic (brown) spots on the leaves, often encircled by a fine yellow halo. Similarly Hagedorn and Inglis (1986) noticed symptoms may first look like as water-soaked spots, which gradually enlarge and dry up, and are often bordered by a narrow yellow or light green zone. Wounds may unite and occasionally abscise, later giving the foliage a ragged appearance. According to Watson (1980), bacterial brown spot leaf symptoms on beans are small, irregular necrotic lesions that are sometimes surrounded by a narrow, pale green chlorotic zone. Lesions may coalesce, dry out and become brittle, giving leaves a tattered appearance.

Morphological and biochemical test results of bacterial brown spot

The characterization of phytopathogenic bacteria helps to know the target pathogen and its biological behaviors. Microscopic observations like shape and grams nature revealed the availability of similar morphological characters among isolates. Growth of bacterial colonies after 48 h under aerobic conditions at 30°C resulted in round, yellow with complete margins, rod shaped and mucoid appearances (Table 2). The gram staining method revealed that test isolates showed rod shaped and gram negative bacteria (Figure 5). KOH test confirmed that all the isolates showed positive reaction and were categorized as gram negative rod shaped phytopathogenic bacteria.

The isolates were negative for oxidase reaction, starch hydrolysis (Figure 7), potato soft rot, hydrogen sulfide (H_2S) production, lactose tests and were positive for KOH (Figure 10), catalase (Figure 11), Aesculine (Figure 6), iron-deficient media (Figure 8), tween 80 hydrolysis

Table 2. Macro and microscopic characteristics of bacterial brown spot isolates.

Number	Isolate code	Colony color	Colony shape	Colony texture	Colony margin	Cell shape
1	keF14	Yellow	Round	Mucoid	Entire	Road
2	keF11	Yellow	Round	Mucoid	Entire	Road
3	Ka14	Yellow	Round	Mucoid	Entire	Road
4	KeF8	Yellow	Round	Mucoid	Entire	Road
5	Tf1	Yellow	Round	Mucoid	Entire	Road
6	KeF1	Yellow	Round	Mucoid	Entire	Road
7	Kaf2	Yellow	Round	Mucoid	Entire	Road
8	KeF5	Yellow	Round	Mucoid	Entire	Road
9	KeF2	Yellow	Round	Mucoid	Entire	Road
10	KeF7	Yellow	Round	Mucoid	Entire	Road
11	Kef6	Yellow	Round	Mucoid	Entire	Road
12	Tf3	Yellow	Round	Mucoid	Entire	Road
13	Ka15	Yellow	Round	Mucoid	Entire	Road
14	KeF13	Yellow	Round	Mucoid	Entire	Road
15	KaF6	Yellow	Round	Mucoid	Entire	Road
16	Tf13	Yellow	Round	Mucoid	Entire	Road
17	Ka10	Yellow	Round	Mucoid	Entire	Road
18	Tdf9	Yellow	Round	Mucoid	Entire	Road

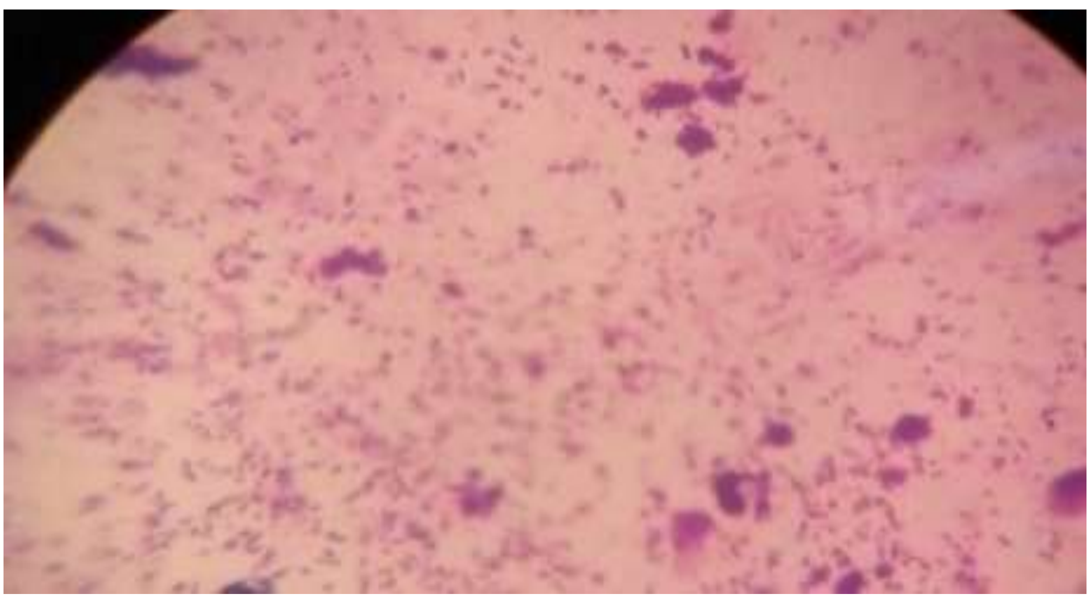

Figure 5. Morphology of rod shaped gram negative bacteria.

(Figure 9), gelatin liquefaction, nitrate reduction, levan production, sucrose, mannitol, sorbitol, maltose, inositol and dulcitol tests (Table 3). Carbon sources were used to distinguish Pss from *P. savastanoi* pv. *Phaseolicola* isolates. Finally, the isolates were identified and considered as *P. syringae* pv. *syringae* on KBC selective media in combination with other biochemical tests conducted to identify the target pathogen.

Consistent with this, Goszczynska et al. (2000) reported that bacterial brown spot isolates are light cream with entire margins, rod and fluorescent. According to Wazeer et al. (2014) growth of bacterial colonies after 72 h under aerobic conditions at 28 ± 2°C resulted round, yellow with complete margins, dome shaped, shiny, smooth and mucoid appearances. Bacterial morphological features alone are of little taxonomic value; because they are too simple to provide enough taxonomic information (Lelliott and Stead, 1987).

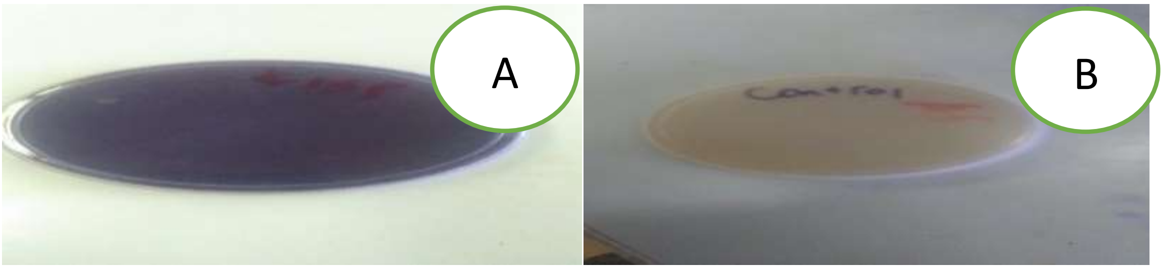

Figure 6. (A&B). Positive for aesculine test and aesculine control on NA.

Figure 7. Negative for Starch test (left) and control (right).

Figure 8. Growth of bacterial brown spot isolate on KB (left) and NA media (right).

Figure 9. Opaque zone around colony recorded as positive reaction for hydrolysis of Tween 80.

Figure 10. Positive for KOH test.

Figure 11. Positive for catalase result.

Similarly, Muedi et al. (2011) stated all fluorescent isolates tested were levan-positive, oxidase -negative, potato soft rot-negative, arginine dihydrolase-negative and mannitol-positive, sorbitol- positive, inositol- positive and carbon sources were used to distinguish Pss from *P. savastanoi* pv. *Phaseolicola* isolates. The result is also in agreement with results obtained by Mohammad (2013) who directed bacterial brown isolates are negative for grams reaction, oxidase reaction, nitrate reduction, starch hydrolysis, lactose, hydrogen sulphide production and positive for gelatin, aesculine hydrolysis, levan production and utilization of carbohydrates.

According to Žarko et al. (2017) report, *P. syringae* pv. *syringae* isolates are positive for levan production, aesculine hydrolysis, gelatin liquefaction and negative for oxidase reaction, potato soft rot, arginine dihydrolase production, tyrosinase activity and tartarate utilization.

Wazeer et al. (2014) reported that *P. syringae* pv. *syringae (*Pss) isolates are positive for mannitol, aesculine, gelatin and negative for arginine and nitrate reduction.

Conclusions

In the present work, field survey, observation and laboratory diagnosis were performed to investigate the status, isolate and identify the causal pathogen. In the present work, we have performed field survey, observation and laboratory diagnosis to investigate the status, isolate and identify the causal pathogen.

In the present work, we have performed field survey, observation and laboratory diagnosis to investigate the

Table 3. Biochemical test results of bacterial brown spot isolates.

		Biochemical tests																		
S/N	***Isolate code***	***KOH***	***H_2O_2***	***Oxidase***	***Starch***	***Potato sr***	***Aesculineeee***	***Nitrate Red[n]***	***Gelatin***	***Lactose***	***Sucrose***	***KB***	***Tween80 Hydrolysis***	***Levan***	***H_2S prd[n]***	***Mannitol***	***Sorbitol***	***Inositol***	***Dulcitol***	***Maltose***
1	*keF14*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
2	*keF11*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
3	*Ka14*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
4	*KeF8*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
5	*Tf1*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
6	*KeF1*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
7	*Kaf2*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
8	*KeF5*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
9	*KeF2*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
10	*KeF7*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
11	*Kef6*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
12	*Tf3*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
13	*Ka15*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
14	*KeF13*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
15	*KaF6*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
16	*Tf13*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
17	*Ka10*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+
18	*Tdf9*	+	+	-	-	-	+	-	+	-	+	+	+	+	-	+	+	+	+	+

status, isolate and identify the causal pathogen bacterial brown spot of mungbean. In Ethiopia, mung bean is a recently introduced crop. It is a niche for several foliar pathogens. Thus, field study conducted to determine the distribution and association of bacterial brown spot disease of mung bean with different biophysical factors and laboratory diagnosis done to isolate and identify the causal pathogens revealed the disease as bacterial brown spot (*P. syringae* pv. *syringae)*. Additional pests like yellow mosaic virus and foliage beetle were observed in combination with the above diseases.

Accordingly, the disease were prevalent and widely distributed. This indicates the favorable environmental conditions coupled with cultivation of susceptible mung bean cultivars and non-implementation of appropriate management practices worsened the problem to maximum. Bacterial brown spot were found as major ones that limits the production of the crop. This implies that appropriate interventions are needed through creating awareness for growers about disease causative agent characteristics, survival, dispersal as well as possible management options to reduce effects below economic injury level. The intensity of disease observed also varied between districts and altitudes. This might be due to differences in weather and management practices applied by farmers during the survey year. This directs worthwhile of conducting similar assessments in different mung bean belt areas of the country for all diseases.

The independent variables such as soil condition, cropping system, growth stage, previous crop and adjacent crop showed association to disease severity with different significance among them. Thus, the disease was favoured by warm to hot semi-arid with mid high land, moist conditions and lush crop canopies. This shows role of weather conditions and agronomic practices are under considerations in the epidemics and severities of the disease advancement. Based on the laboratory diagnosis a newly emerging disease of mung bean named as bacterial brown spot of mung bean (*P. syringae* pv. *syringae)* were identified. This implies more isolates covering wide agro-ecologies shall be collected and identified further. Result from pathogenicity test revealed that all tested isolates were symptomatic to the inoculated susceptible variety. The production of typical symptoms for bacterial brown spot was observed under greenhouse condition of controlled environment. This implies that there is an urgent need to study the aggressiveness of isolates of the crop.

Recommendations

This study showed the current status of major mung bean disease in the study areas,identified to be bacterial foliar disease in growing areas of North Shewa and South Wollo *zones* of Ethiopia. At present, this disease threatens mung bean production.
Therefore, based on this study the following are suggested or recommended:

(i) Survey should be undertaken to know the distribution of this disease and other pests across the producing regions of Ethiopia.
(ii) Role of weather conditions in the epidemics and severities of the disease may be investigated through correlations and environmental models as it was not much studied in the present study.
(iii) This study could provide first information on morphological and biochemical characterization of bacterial brown spot of mung bean in North Shewa and South Wollo growing *zones* of Ethiopia and it is useful for researchers to go on further molecular characterization studies of isolates.
(iv) It would be more interesting to study phenotypic characteristics of a large population of newly emerged Pss from various host plants in different regions of Ethiopia. Further collection, isolating and identification work should be sustained.
(v) Awareness creation about diseases is very necessary for farmers, development agents and others commercial producers since the problem is not well popular and understood by the mung bean producers and partners.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Ali MZ, Khan MAA, Rahaman AKMM, Ahmed M, Ahsan AFMS (2010). Study on seed quality and performance of some mung bean varieties in Bangladesh. International Journal Experiment Agriculture 1(2):10-15.

Altieri MA, Ponti L, Nicholls C (2005). Enhanced pest management through soil health: toward a belowground habitat management strategy. University of California, Berkeley. Bio-dynamics, pp. 33-40.

Aneja KR (2003). Experiment in Plant Pathology, Microbiology and Biotechnology. New Age International (p) Ltd., Publishers, New Delhi, pp. 74- 275.

Aneja R (1996). Experiments in Microbiology, Plant pathology, Tissue Culture and Mushroom Cultivation. Wishwa Prakashan, New Age International (P) Limited, New Delhi.

Ashrar M, Siririnives P, Sadiq MS, Saleem M (2001). AVRDC Germplasm: Its utilization and development of improved mungbean.Pakistan Journal of Botany, p. 33.

Asrat, Fekadu G., Alemayehu F, Yeyis R (2012). Analysis of Multi-environment Grain Yield Trials in Mung Bean (*Vigna radiate* L.) Wilczek Based on GGE Bipot in Southern Ethiopia. Journal of Agricultural Science Technology 14:389-398.

BOA (Bureau of Agriculture) (2000). Amhara National Regional State (ANRS) Bureau of Agriculture, Special report on technology packages. BOA, Bahir Dar, Ethiopia, pp. 4-6.

Chadha ML (2010). Short duration mung bean: a new success in South Asia. Asia-Pacific association of agricultural research institutions: Thailand. Bangkok FAO RAP. pp. 2-55.

Chemeda F (2003). Relation between common bacterial blight severity and bean yield loss in pure stand and bean-maize intercropping systems. International Journal of Pest Management 49(3):177-185.

CSA (Central Statistical Agency) (2017). Report on Area and Production of Major Crops. Volume 1. Addis Ababa, Ethiopia.

Das S, Shekhar UD, Ghosh P (2014). Assessment of molecular genetic diversity in some green gram cultivars as evealed by ISSR analysis. Advances in Applied Science Research 5(2):93-97.

Dickey RS, Kelman A (1988). '*Caratovora'* or soft rot group. In: Laboratory guide for identification of plant pathogenic bacteria 2nd ed. (ed. N.W. Schaad.). APS Press St. paul, Minnesota. pp. 81-84.

ECXA (2014). Ethiopia Commodity Exchange Rings Bell for mung bean Addis Ababa.

EPP (Ethiopian Pulses Profile) (2004). "Ethiopian export promotion agency, product development and market research directorate, May 2004 Addis Ababa, Ethiopia.

Fahy P, Hayward A (1983). Media and methods. Plant bacterial disease: A diagnostic guide. Academic, New York. pp. 337-378.

FAO Food and Agricultural Organization of the United Nations) (2017). FAO/INFOODS global food composition database for pulses. Rome.

Firdous SS, Asghar R, Ul-Haque MI, Waheed A, Afzal SN, Mirza MY (2009). Pathogenesis of *Pseudomonas syringae* pv. *sesame* associated with sesame (*Sesamum indicum* L.) bacterial leaf spot. Pakistan Journal of Botany 41(2):927-934.

Geiser DM, Lewis Ivey ML, Hakiza G, Juba JH, Miller SA (2005). *Gbberella xylariodes* (anamorph: *Fusarium xylarioides*), a causative agent of coffee wilt disease in Africa, is a previously unrecognized member of the G. fujikuroi species complex. Mycologia 97:191-201.

Goszczynska T, Serfontein JJ, Serfontein S (2000). Introduction to practical phytobacteriology. A manual for phytobacteriology. South Africa. https://www.passeidireto.com/arquivo/59976661/introduction-to-practical-phytobacteriology

Grewal JS (1987). Diseases of mung bean in India. In: Proceedings of First International Mung Bean Symposium, 16-19 Aug, 1977. University of Philippines, Los Banos pp. 165-168.

Habtamu T, Chemeda F, Samuel S,Kindie T (2015). Effect of integrated cultural practices on the epidemics of chocolate spot (*Botrytis fabae*) of faba bean (*Vicia faba* L) in Hararghe Highlands, Ethiopia. Global Journal of Pests, Diseases and Crop Protection 3(4):113-123.

Hagedorn DJ, Inglis DA (1986). Handbook of Bean Diseases. Madison, WI: University of Wisconsin.

Hildebrand DC (1998). Pectate and pectin gel for differentiation of *Pseudomonas* sp. and other bacterial plant pathogens. Phytopathology 61:1430-439.

Howard FS, David HG, Gary DF, Robert MH (2016). High Plains Pest Management. Submitted to University of Nebraska, Western Nebraska.

Ignjatov M, Miloševiæ M, Nikoliæ Z, Vujakoviæ M, Petroviæ D (2007). Characterization of *Pseudomonas savastanoi* pv. *glycinea* isolates from Vojvodina. [Available on internet] http:// www.au.poznan.pl/ptfit/pdf/pp45/ pp_45_05.pdf [Access date: 02/08/2009].

Imrie BC, Lawn RJ (1991). Mung bean: The Australian Experience, Proc. 1st Australian mung bean Work-Shop, Bribane P. 53.

Itefa D (2016). General Characteristics and Genetic Improvement Status of Mung Bean (*Vigna radiata* L.) in Ethiopia. International Journal of Agriculture Innovations and Research 5(2):2319-1473.

Jamadar MM (1988). Studies on leaf spot of green gram (Vigna radiata L.) Wilczek caused by Cercospora canescens. MSc. Thesis, University of Agricultural Scicience, Dharwad

Jitendra K, Anila D (2018). Severity of Bacterial Leaf Spot (*Xanthomonas axonopodis* pv. *vignaradiatae*) of Green Gram in DifferentTehsils of Udaipur District, Rajasthan, India International Journal of Current Microbiology and Applied Sciences 7(3):2924-2931. doi:https://doi.org/10.20546/ijcmas.2018.703.338.

Kalu District Office of Agriculture (KDOA) (2010). Annual Report. Kombolcha, Ethiopia.

Karthikeyan A, Shobhana VG, Sudha M, Raveendran M, Senthil N, Pandiyan M, Nagarajan P (2014). Mung bean yellow mosaic virus (MYMV): a threat to green gram (*Vigna radiata*) production in Asia. International Journal of Pest Management 60(4):314-324, DOI: 10.1080/09670874.2014.982230.

Kavyashree MC (2014). Studies on Fungal Foliar Diseases of Green gram *Vigna Radiata* (L.) Wilcze. MSc. Thesis, University of Agricultural Sciences, Dharwad.

Khan A, Malik, MA (2001). Determining biological yield potential of different mung bean cultivars. On line Journal of Biological Science 1(7):575-576.

Khan MA, Naveed K, Ali K, Ahmad B, Jan S (2012). Impact of mung bean-maize intercropping on growth and yield of mung bean. Pakistan Journal of Weed Science Research 18(2):191-200.

Khattak GSS, Haq MA, Ashraf M, Mcneilly T (2001). Genetic basis of variation of yield, and yield components in mung bean (*Vigna radiata* (L.) Wilczek). Hereditas 134:211-217. Lund, Sweden.

Kijana R, Abang M, Edema R, Mukankusi C, Buruchara R (2017). Prevalence of Angular Leaf Spot Disease and Sources of Resistance in Common Bean in Eastern Democratic Republic of Congo. African Crop Science Journal 25(1):109-122.

Klement Z (1990). Inoculation of plant tissues. Cancer and dieback disease. In: Methods in phytobacteriology, eds. by Z. Klement, K. Rudolph and D. C. Sands, pp. 105-106. Akademiai Kiado, Budapest, Hungary.

Kumari R, Shekhawat KS, Gupta R, Khokhar MK (2012). Integrated management against root- rot of mung bean (*Vigna radiata* (L.) Wilczek) incited by *macrophomina phaseolina.* Journal of Plant Pathology and Microbiology 3:5.

Lambrides C, Godwin ID (2007). Mung Bean Genome Mapping and Molecular Breeding in Plants, Pulses, Sugar and Tuber Crops, P. 3.

Lelliott RA, Stead DE (1987). Methods for the Diagnosis of Bacterial Diseases of Plants. Blackwell Scientific Publications, London, UK. ISBN-13: 9780632012336, P. 216.

Makkouk KM, Kumari SG, Huges JDA, Muniyappa V, Kulkarni NK (2003). Other legumes. In: Virus and virus luke disease of major crops in developing countries, (Eds: Leobension D. and Thollapilly G.), Klieur Acadmaic Publisher, pp. 447-476.

Minh NP (2014). Different factors affecting to mung bean (*Phaseolus aureus*) tofu production. International Journal of Multidisciplinary Research and Development 1(4):105-110.

Mohammad G (2013). Identification of Bacterial Agent of the Bark Canker of Peach in the Province of Ilam. Current Research in Bacteriology 6(1):1-4.

Muedi HTH, Fourie D, McLaren NW (2015). Distribution and severity of bacterial brown spot on dry beans in South Africa: An update. South Africa Journal of Science 111(3/4).

Muedi HTH, Fourie D, McLaren NW (2011). Characterization of Bacterial Brown Spot Pathogen From Dry Bean Production Areas Of South Africa. African Crop Science Journal 19(4):357-367.

Mwangombe A, Wagara I, Kimenju J, Buruchara R (2007). Occurrence and severity of angular leaf spot of common bean in Kenya as influenced by geographical location, altitude and agro-ecological zones. Plant Pathology Journal 6(3):235-241.

Ngugi HK, King SB, Abayo GO, Reddy YVR (2002). Prevalence, incidence and severity of sorghum diseases in Western Kenya. Plant Disease 86:65-70.

Njingulula P, Wimba M, Musakamba M, Masukim KF, Katafire M, Ugen M (2014). Strengthening local seed systems within the bean value chain experience of agricultural innovation platforms in the democratic republic of Congo. African Crop Science Journal 22:1003-1012.

Pursglove JW (2003). Tropical Crops. Longman, London.

Robert MH (2009). Bacterial brown spot of dry beans in Nebraska. University of Nebraska. http://extensionpublications.unl.edu/assets/pdf/g1957.pdf

Sadeghipour O (2009). The influence of water stress on biomass and harvest index in three mung bean (*Vigna radiata* (L.) Wilczek) cultivars. Asian Journal Plant Science 8:245-249.

Saettler AW (2005). Diseases caused by bacteria. Compendium of bean diseases. 2nd ed. St. Paul, MN: American Phytopathological Society pp. 46-47.

Sands DC (1990). Physiological Criteria Determinative tests. In: Klement Z, Rudolph K, Sands DC (eds.) Methods in Phytobacteriology, Akademiai Kiade, Budapast, Hungary.

Sands DC (1999). Physiological criteria Determinative test. Methods in phytobacteriology. Akademia Kiado, Budapest, Hungary, pp. 137-143.

Schaad NW (1980). Laboratory guide for the identification of plant pathogenic bacteria. St. Paul (Minn.). American Phytopathological Society, pp. 28-45.

Scheuermann KK, Raimondi JV, Marschalek R, Andrade A, Wickert E (2012). *Magnapor theoryzae* genetic diversity and its outcomes on the search for durable resistance.In: Mahmut Caliskan (ed.). ISBN 978-953-51-0157- 4. The Molecular Basis of Plant Genetic Diversity, Chapter, 15:331-356.

Schwartz HF, Brick MA, Harveson RM, Franc GD (2019). Bacterial Diseases of Beans. Bullenti 562A, Colorado State University Extension. Fort Collins.

Serfontein JJ (1994). Occurrence of bacterial brown spot of dry beans in the Transvaal Province of South Africa. Plant Pathology 43:597-599.

Sseruwagi P, Sserubombweb WS, Legga DJP, Ndunguruc J, Threshd JM (2004). Methods of surveying the incidence and severity of cassava mosaic disease and whitefly vector populations on cassava in Africa: A review. Virus Research, pp. 129-142.

Stenglein S, Ploper LD, Vizgarra O, Balatt P (2003). Angular leaf spot: a disease caused by the fungus *Phaeoisariopsis griseola* (Sacc.) Ferraris on *Phaseolus vulgaris* L. Advances of Applied Microbiology 52:209-243.

Suli S, Ye Z, Zhendong Z, Jing J, Canxing D, Xiaofei W, Wang X (2017). An Emerging Disease Caused by *Pseudomonas syringae* pv. *Phaseolicola* Threatens Mung Bean Production in China. The American Phytopathological Society. Plant Disease 101:95-102.

Summerell BA, Gunn LV, Bullock S, Tesoriero LT, Burgess LW (2006). Vascular wilt of basin in Australia. Australasian Plant Pathology 35:65-67.

Teame G, Ephrem S, Lemma D, Getachew B (2017). Adaptation Study of Mung Bean (*Vigna radiate*) Varieties in Raya Valley, Northern Ethiopia. Current Research in Agricultural Sciences 4(4):91-95.

Tensay A (2015). MungBean (*Vigna radiata* (L.) Wilczek) (Fabaceae) Landrace Diversity in Ethiopia. Addis Ababa, Ethiopia. http://etd.aau.edu.et/bitstream/handle/123456789/9376/Tensay%20Ayalew.pdf?sequence=1&isAllowed=y

Wachenje CW (2002). Bean production constraints, bean seed quality and effect of intercropping on floury leaf spot disease and yields in Taita Taveta district, Kenya. MSc Thesis, University of Nairobi 117p.

Ocimati W, Were E, Jeroen CJG, Pablo T, Gloria VN, Guy B (2018). Risks Posed by Intercrops and Weeds as Alternative Hosts to Xanthomonas campestris pv. musacearum in Banana Fields. Frontiers in Plant Science 9:1471. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6192450/

Waniale A, Talwana H, Wanyera N (2012). Agro-morphological characterization of exotic and local green gram mung bean (*Vigna radiata*) for breeding purposes in Uganda, P. 28.

Watson DRW (1980). Identification of bacterial brown spot of bean in New Zealand. New Zealand Journal of Agricultural Research 23:267-272.

Wazeer AH, Farida F N, Jivan MM, Al D (2014). First Record of *Pseudomonas syringae* pv. *syringae* in Iraq using Conventional and Specific PCR Protocol. IOSR Journal of Agriculture and Veterinary Science 7:01-08.

Wheeler BEJ (1969). An Introduction to Plant Disease. John Wiley Sons Ltd., London P. 301.

York MK, Taylor MM, Hardy J, Henry M (2004). Biochemical tests for the identification of aerobic bacteria, Clinical Microbiology Procedures Handbook, (2nd Ed.). ASM Press, Washington, DC 3:17-39.

Žarko I, Tatjana P, Tatjana P, Jovana B, Nenad T, Snježana H (2017). Characterization of *Pseudomonas syringae* pv. *syringae,* Causal Agent of Citrus Blast of Mandarin in Montenegro. Plant Pathology Journal 33(1):21-33.

Zewde T, Chemeda F, Sakhuja PK, Seid A (2007). Association of white rot (*Sclerotium cepivorum*) of garlic with environmental factors and cultural practice in the North Shewa highlands of Ethiopia. Crop Protection 26(10):1566-1573.

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Agro-morphological response of some groundnut genotypes (*Arachis hypogaea* L.) in water deficit conditions

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Groundnut is a crop that can be grown under varied production conditions (in intercropping or rotation with cereals). In Niger, the production of groundnut is decreasing over the year due to drought and low soil nutrients. In this work, an agro-morphological evaluation of five groundnut genotypes (55-437, ICG12697, ICG4750, JL24 and ICG8751) under water deficit was carried out in order to identify the best performing genotypes for seed and forage production. Intermittent water deficit was imposed from the 53rd day after sowing, which was the beginning of pod filling. The other plants were well watered until harvest. The results showed that in water stress conditions all yield parameters (pod number, pod weight, seed number, seed weight, harvest index and pod filling rate) and vegetative parameters (aerial biomass and height) with the exception of the number of branches decreased. The principal component analysis revealed that genotypes with a good harvest index and high pod filling rate have good seed yield. Therefore, harvest index and filling rate can be used for the selection of genotypes under water stress conditions. The genotypes 55-437, ICG4750 and ICG12697 proved to be the best performers under water stress and well-watered conditions. These ones accumulate vegetative biomass as proportionate way to the production and filling of the seeds in contrast to JL24 and ICG8751, which tend to accumulate vegetative biomass to the detriment of the production and filling of pods.

Key words: Agromorphologic, yield, groundnut, water deficit, Niger.

INTRODUCTION

In Niger, the agricultural farming system is mainly rainfed. The dominant cropping system is the association of cereals and legume species with a predominance of millet, cowpeas, and groundnut. Groundnut (*Arachis hypogaea* L.) is the main legume crop grown after cowpeas. Cultivated in association or in rotation with millet, it is of socio-economic and ecological importance to small farmers.

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Table 1. Origin and earliness of groundnut genotypes used.

Name	Origin	Response to drought
ICG 12697	India	Tolerant
ICG 8751	Perou	Sensible
JL 24	India	Sensible
55-437	Senegal	Tolerant
ICG 4750	Paraguay	Tolerant

However, its production is low, 302,524 tons in 2014 (RECA, 2015). One of the most limiting factors in agriculture is water deficit (FAO, 2014) due to the large inter-annual climate variations such as rainfall variations which cause drought (Himeno et al., 2009). Drought stress impacts plant growth at many different levels. At the physiological level water deficit is perceived in roots and results in turgor loss, reduced water potential and decreased stomata conductance (Zhang and Uwe, 2017). The impact of drought on groundnut yield depends on the intensity of water stress and the stage of its appearance. The reduction in pod production by water stress is higher in the flowering stage than during the pod filling stage (Halilou, 2016). Among the most relevant mechanisms of drought tolerance in groundnut are root development and stomatal regulation (Halilou, 2016), while others such as the accumulation of abscissic acid (ABA) or proline appear to present less of interest (Madhusudhan et al., 2002).

The short cycle groundnut (70 to 90 days) uses the drought escape mechanism, which is particularly effective in environment with frequent water deficit at the end of the cycle (Clavel et al., 2007). This mechanism allows them to avoid end-of-cycle dryness, which corresponds to the very sensitive pod filling stage in groundnut. Genotypic variations in seed yield under intermittent drought have been observed in groundnut (Halilou, 2015). This work aimed to assess the effect of intermittent water stress at the end of the cycle on the yield of five groundnut genotypes in order to determine the most relevant traits in the expression of yield.

MATERIALS AND METHODS

Plant material

Five groundnut genotypes were chosen for this experiment based on their response to drought (Table 1). The seeds used were made available to us by the International Research Institute for Crops of the Semi-Arid Tropics (ICRISAT) in Sadoré (Niger). All five genotypes have a 90-day development cycle.

Experimental conditions

The trial was conducted in pots during the 2016 rainy season (July-October) at the ICRISAT station in Sadoré (latitude 13° 15'N and longitude 2° 18'E.) located 45 km southwest of Niamey in Niger. The experimental plants were grown in pots stored on tarpaulin support to prevent root contact with the soil. The trial was put in natural conditions of lighting, temperature, and humidity. The pots were filled with sandy soil deficient in phosphorus taken from the surface horizon (20 cm deep) of field 8°C at the Sadoré station. The 35 l plastic pots are filled with 34 kg of soil enriched with manure (30 g.kg^{-1} soil). The bottom of each jar has been pre-drilled to let the water drip out. Sowing was carried out on July 15, 2016, at the rate of three seeds per pot followed by thinned to one plant per pot 16 days after sowing. During the imposition of stress, the plants were protected from rainwater by a mobile shed with a translucent roof. Climatic data (temperature and humidity) were recorded daily using a thermo hygrometer (Tiny tag Ultra 2 TGU-4500 Gemini Data loggers Ltd, Chichester, UK) installed next to the trial. During the trial, the average temperature was 29°C while the relative humidity was 75% (Figure 1).

Experimental device

The experimental design was a split plot in randomized blocks with four repetitions. Two factors were studied: the two-levels water regime and the five-levels genotypes. Each block is made up of 20 pots including 5 pots per repetition. Each water regime is applied to plants in the same block. The two levels of water regimes are: T0: well-watered; T1: suspend watering at pod filling stage 53 days after sowing (DAS) for 9 days.

Measured parameters

The following phenological stages were recorded: emergence, date of start of flowering, date of start of pod filling. These parameters were measured on all the pots for the two treatments. The stage was noted when 50% of the plants in the block have reached the stage. At maturity, the following parameters per plant were measured: total height, number of twigs, pods and seeds. After 8 days of drying in the greenhouse, the dry biomass of the tops, pods, seeds and cockles was determined.

The pod filling rate (TR) was calculated by the formula: TR = seed weight / pod weight.

The pod harvest index (IR) was calculated using the following formula:

$$IR = 1.65 \times \text{pod weight} / (\text{pod weight} \times 1.65 + \text{aerial biomass weight})$$

The correction coefficient 1.65 was used to adjust for differences in the energy requirements of the peanut to produce the dry matter of the pods compared to the vegetative part (Duncan et al., 1978).

Data analysis

The analysis of variance was carried out using the Minitab16

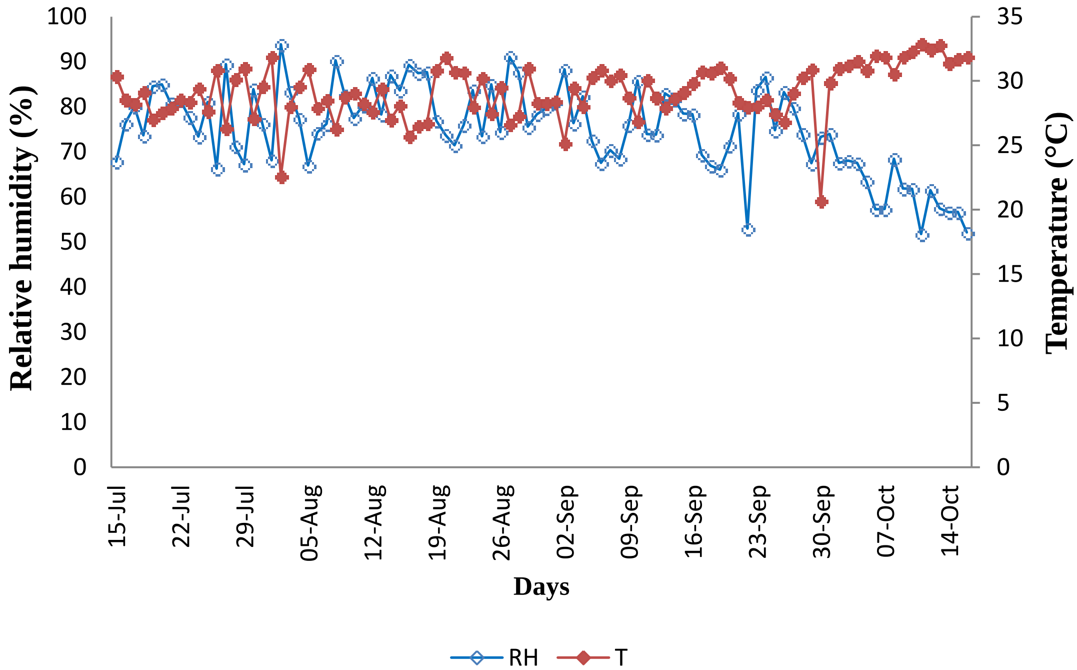

Figure 1. Variation in temperature (T) and relative humidity (RH) during the test period.

software. The separation of the means for the various measured parameters was carried out by the Tukey test at the threshold of α = 5%. The significance of the correlation between the parameters studied was verified using the Pearson correlation test. The principal component analysis (PCA) was carried out to choose the most relevant parameters, which allow the genotypes to be discriminated.

RESULTS

Phenology

All genotypes emerged on average after 5 days after sowing (DAS) (Table 2). There are no significant differences between the genotypes for the start of flowering and pod filling dates, which occurred, on average at 25 and 53 DAS, respectively.

Influence of water deficit on growth parameters and groundnut yield

The results show that when the plants are well watered (T0), there are no significant differences (p> 0.05) between the genotypes for the total height of the plant, the number of branches, the yield pods, empty pod, and aerial biomass as well as the pod filling rate (Table 3). However, significant differences exist between the genotypes for the other parameters. The Tukey test made it possible to separate the genotypes into two groups for the number of pods / plants: 55-437, ICG12697 and ICG8751 produced more pods than the other genotypes.

Genotype 55-437 produced the best number of seeds/plant (95.25 seeds / plant), followed by ICG12697 (89 seeds/plant). JL24 produced the lowest number of seeds (56.75 seeds / plant). This genotype also produced the lowest seed/plant weight (18.84 g) and pod harvest index (49.75%) compared to other genotypes that have similar values.

When plants were subjected to water stress (T1), the results indicate that there were no significant differences between the genotypes for the total height of the plant, the number of pod per plant, the yield of pods, seeds and hulls (Table 3). However, significant differences exist for the number of pods (p <0.01). The best number was obtained for ICG12697 and ICG4750, with approximately 30 pods / plant and the lowest by ICG8751 and JL24 (19 pods / plant). The best number of seeds / plant was recorded for 55-437, ICG12697 and ICG4750 compared to genotypes ICG8751 and JL24. Stopping watering for 9 days resulted in an average reduction of 45.15% in the number of pods and 55% in the number of seeds compared to well-watered. Genotype 55-437 has the highest pod harvest index (50.33%) and ICG8751 the lowest (35.21%). The other genotypes (JL24, ICG4750 and ICG12697) have an intermediate index. The induction of water stress results in a variable reduction in haulm yield depending on the genotypes (Figure 2). This reduction was greater for 55-437 and JL24 (39%) and to some extent ICG12697 (35%). It was lower for ICG4750 (29%) and ICG8751 (24%) (Figure 3).4

The reduction in yield due to stress was even more

Table 2. Phenological stages of genotypes studied (in number of days after sowing).

Genotypes	Emergence	Beginning of flowering	Beginning of pod filling
55-437	5.00	24.75	52.75
ICG12697	5.13	24.88	53.00
ICG4750	5.13	25.00	52.13
ICG8751	5.13	25.75	53.50
JL24	5.63	26.00	53.75
SE±	0.32	1.12	1.66
Significance	ns	ns	ns

Table 3. Total height of the plant, yield and its components per plant for five peanut genotypes well-watered (T0) and subjected to water deficit (T1).

Treat	Genotypes	Ht (cm)	NRm	Ngo	Ngr	Pgo (g)	Pgr (g)	Pcq (g)	P Fanes (g)	TR(%)	IR(%)
	55-437	35.25	7.75	53.50^{a}	95.25^{a}	35.65	28.8^{a}	7.73	36.33	81.27	75.08^{a}
	ICG12697	38.00	9.00	50.75^{a}	89.00^{b}	37.59	27.27^{a}	10.31	39.60	72.39	67.18^{a}
	ICG4750	36.75	8.50	39.50^{b}	76.25^{c}	37.27	28.42^{a}	9.63	38.47	72.53	67.09^{a}
	ICG8751	37.00	10.25	48.50^{a}	79.50^{c}	34.35	24.22^{ab}	9.54	39.26	70.68	62.86^{ab}
T0	JL24	35.65	11.00	38.50^{b}	56.75d	26.89	18.84^{b}	7.13	44.09	70.25	49.75^{b}
	SE±	4.11	1.87	4.12	4.14	5.00	2.68	1.67	2.54	5.60	6.99
	Significance	ns	ns	**	***	ns	**	ns	ns	Ns	**
	55-437	35.00	11	27.00^{b}	50.75^{a}	17.89	13.49	3.70	22.16	74.99	50.33^{a}
	ICG12697	33.50	12	29.25^{ab}	49.25^{a}	22.70	13.58	4.63	25.88	73.03	46.44^{ab}
	ICG4750	34.25	13.25	32.75^{a}	58.00^{a}	17.61	16.55	5.08	27.33	75.08	49.50^{ab}
	ICG8751	35.50	13.5	19.00^{c}	33.50^{b}	18.72	10.05	5.03	29.89	62.76	35.21^{b}
	JL24	35.25	12	19.25^{c}	39.00^{b}	15.42	11.48	4.52	26.74	64.67	43.13^{ab}
T1	**SE±**	5.736	2.604	2.48	4.31	5.42	4.067	1.92	4.155	8.89	7.49
	Significance	ns	ns	***	***	0,74	ns	ns	ns	Ns	*
	Genotype	ns	ns	***	***	ns	ns	ns	*	**	**
	Treatement (T)	ns	**	***	***	***	***	***	***	Ns	**
	Geno*T	ns	ns	***	**	ns	ns	ns	ns	Ns	ns

*, **, *** = significant at the probability threshold of 0.05, 0.01 and 0.00 respectively; ns = not significant (p> 0.05). The figures bearing the same letter (s) in the same column are not significantly different at the threshold of p <0.05. Ht: Total height of the plant; NRm: Number of branches; Ngo: Number of pods; Ngr: Number of seeds; Pgo: Weight of pods; Pgr: Weight of seeds; Pcq: Weight of the empty pod; Fanes: Weight of aerial biomass; TR: Pod filling rate; IR: Pod harvest index.

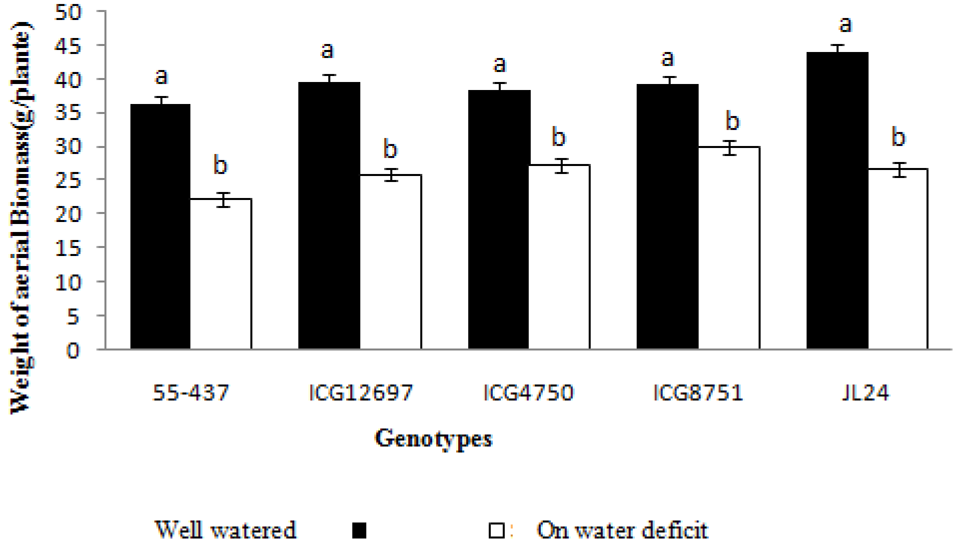

Figure 2. Comparison of the aerial biomass yield of five groundnut genotypes subjected to two water regimes (well-watered and under stress).

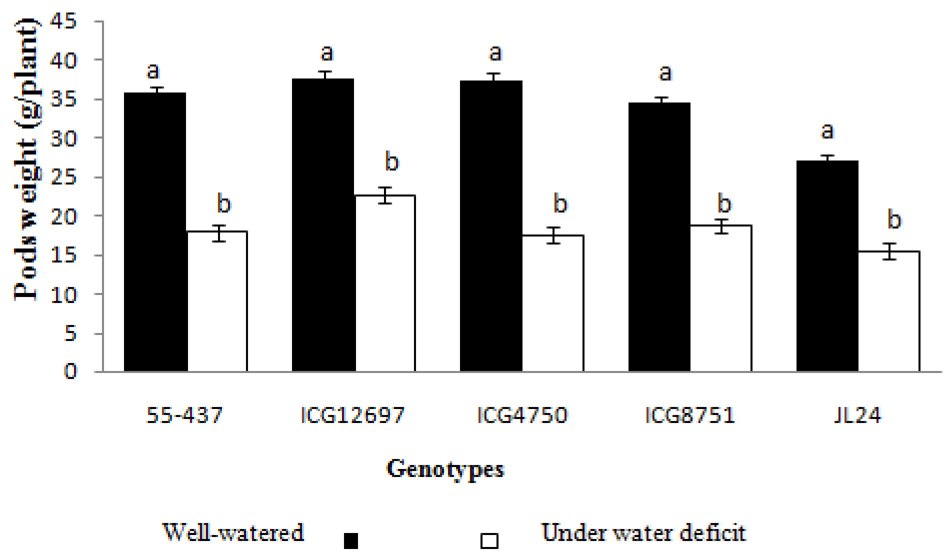

Figure 3. Comparison of pod yield of five groundnut genotypes subjected to two water regimes (well watered and under stress).

important for seeds and pods. The reduction in pod yield was around 50% for 55-437 and ICG4750, 46% for ICG8751, 43% for JL24 and 40% for ICG12697 (Figure 4). The reduction in seed yield (Figure 4) is greater than or equal to 50% for three genotypes: ICG8751 (59%), 55-437 (53%) and ICG12697 (50%). The reduction is around 40% for the other two genotypes (ICG8751 and JL24).

Correlation between the measured parameters

Analysis of the correlation matrix under well water conditions (Table 3) shows significant negative correlations between some vegetative parameters and yield. Thus, the number of branches/plant (NB) was negatively and significantly correlated with the weight of seeds/plant (r^2 = -0.94) and the harvest index (r^2= -0.95). Yield parameters such as, weight of seeds/plant (WS), number of seeds/plant (NS) and harvest index were negatively correlated with aerial biomass yield, r^2 = -0.93; r^2 = -0.90; r^2 = -0.97respectively.

Apart from the negative correlations between the vegetative parameters and the yield parameters. There were positive correlations between the vegetative

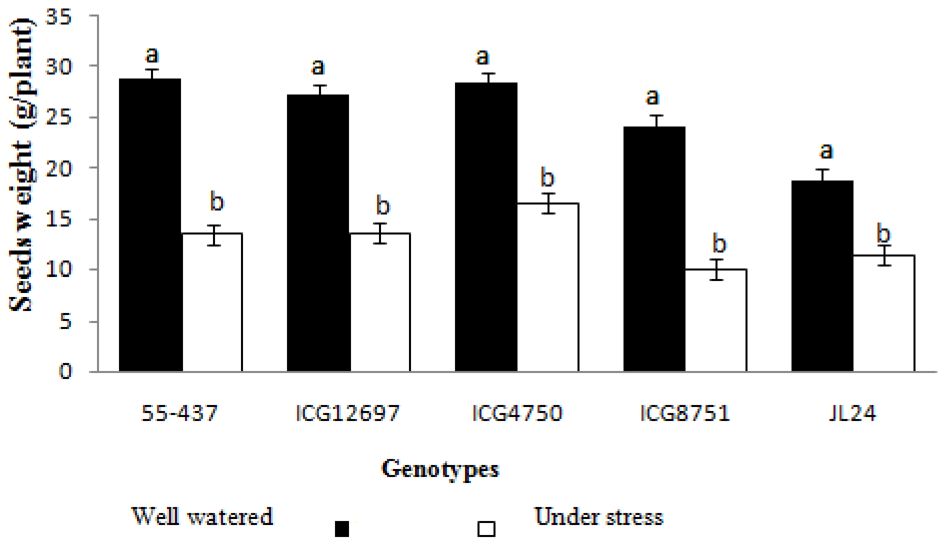

Figure 4. Comparison of the seed yield of five groundnut genotypes subjected to two water regimes (well watered and under stress).

parameters on the one hand and between the yield parameters on the other hand.

Indeed the number of branch/plant (NB) was positively correlated with the aerial biomass yield (r^2 = 0.89). A positive and significant correlation also exists between the weight of seeds/plant and the pod harvest index (r^2 = 0.94) on the one hand and between the number of pods / plant and that of seeds / plant on the other hand (r^2= 0.89) (Table 4).

Under water stress conditions (Table 5), there is no significant correlation between the vegetative parameters and the yield parameters. However, there are significant positive correlations between the vegetative parameters on the one hand and the yield parameters on the other. Note that under water stress the correlation between the weight of seeds/plant and the pod harvest index is not significant.

Eigenvalues and contributions of the characters to the axes of the principal component analysis for the two water regimes

The sum of the proportions of the eigenvalues of the axes of the PCA shows that the first two axes concentrate 91.8% of the information under well-watered conditions, and 86% for the plants under water deficit (Table 6). The first two axes can therefore guarantee precise analyzes. In non-limiting water supply conditions (Figure 5), axis 1 concentrates 67% of the information. It contrasts the vegetative parameters (aerial biomass and number of branches) with the yield parameters (Pgo, Ngr, Pgr, and IR). Axis 2 contrasts the height of plants and weight of empty pods with the pod filling rate. Axis 1 can be defined as the parameters axis explaining the expression of yields and axis 2 for the growth parameters.

In water deficit conditions (Figure 6), axis 1 concentrates 58.8% of the information and opposes the yield parameters (Ngo, Ngr, Pgr, TR and IR) to certain vegetative parameters (aerial biomass yield and plant height). This axis can also be defined as the axis of the parameters participating in the expression of the yields. Axis 2 opposes the aerial biomass, the pod weight and the number of pod to the plant height. This axis can be defined as the axis of growth parameters.

DISCUSSION

The results of the study showed that yields decreased under water deficit conditions compared to the well-watered. There was no significant difference in seed yield for genotypes under water deficit conditions. Among the genotypes studied, 55-437, ICG12697, and ICG4750 gave the best seed yields under water deficit and well-watered conditions. These genotypes also gave the best seeds harvest index and seeds filling rates.

Our results also show that the number of pods is more affected by water stress than the number of seeds. These results corroborate those of Nassar et al. (2018) on 20 peanut genotypes. According to Sharma and Sivakumar (1991), the decrease in the number of pods/plant under water stress is due to the compaction of the soil, which affects their development. Dahanayake et al. (2015) explain this reduction by the abortion of flowers, or due to abortion of newly formed seed (Vurayai et al., 2011).

There was a significant correlation in the harvest index for the seed weight of the well-watered plants, and not

Table 4. Correlation matrix between the parameters measured for plants normally supplied with water (T0).

Paramèter	Ht	NRm	Ngo	Pgo	Ngr	Pgr	Pcq	Fanes	TR	IR
Ht	1									
NRm	0.05									
Ngo	0.13	-0.51	1							
Pgo	0.54	-0.78	0.52	1						
Ngr	0.20	-0.82	**0.89**	0.8	1.0					
Pgr	0.23	**-0.94**	0.54	**0.9**	0.9	1				
Pcq	**0.93**	-0.22	0.22	0.8	0.4	0.52	1			
Fanes	-0.01	**0.89**	-0.67	-0.8	**-0.9**	**-0.93**	-0.33	1		
TR	-0.52	-0.81	0.63	0.4	0.7	0.62	-0.30	-0.74	1	
Ir	0.01	**-0.95**	0.72	0.8	**0.9**	**0.94**	0.29	**-0.97**	0.82	1

In bold, significant values (except diagonal) at the alpha threshold = 0.050 (bilateral test); Ht: Total height of the plant; NRm: Number of Branchs / plant; Ngo: Number of pods / plant; Pgo: Weight of pods / plant; Ngr: Number of seeds / plant; Pgr: Weight of seeds / plant; Pcq: Empty pods weight/ plant; Fanes: Aerial biomass; TR: Pod filling rate, IR: Pod harvest index.

Table 5. Correlation matrix between the parameters measured for plants under stress (T1).

Paramèter	Ht	NRm	Ngo	Pgo	Ngr	Pgr	Pcq	Fanes	TR	IR
Ht	1									
NRm	0.10	1								
Ngo	-0.80	-0.08	1							
Pgo	-0.73	-0.01	0.40	1						
Ngr	-0.68	-0.22	**0.97**	0.18	1					
Pgr	-0.66	-0.05	**0.95**	0.12	**0.98**	1				
Pcq	-0.12	**0.95**	0.00	0.10	-0.15	0.02	1			
Fanes	0.21	**0.93**	-0.38	-0.02	-0.52	-0.36	**0.92**	1		
TR	-0.68	-0.40	**0.94**	0.32	**0.96**	**0.89**	-0.35	-0.68	1	
IR	-0.52	-0.58	0.78	0.00	**0.90**	0.84	-0.50	-0.79	**0.91**	1

significant for the plants under water deficit. However, there was no significant correlation between the harvest index and the pod weight for the two treatments. Our results were in contradiction with those of Halilou (2016) who found a strong correlation between the harvest index and the pod yield. Our results were explained by the fact that some genotypes (ICG8751 and JL24) under water deficit and well-watered conditions tend to produce more biomass than pod. Groundnut yields cannot be explained by pod weight alone, but by seed size and pod filling rate. Otherwise, the ability of plants to transfer assimilates from the vegetative system to pods (Bennett et al., 2012).

Table 6. Eigenvalues and contributions of the characters to the axes of the principal component analysis for the two treatments.

Treatment T0			**Treatment T1**	
	Axis1	**Axis2**	**Axis1**	**Axis2**
Eigenvalues	6.69	2.47	5.87	2.72
Proportion (%)	67	24.8	58.8	27.2
Accumulation (%)	67	91.8	58.8	86
		Correlation between variables and axes		
Ht	0.068	**-0.614**	-0.28	-0.29
NRm	**-0.355**	-0.13	-0.204	**0.495**
Ngo	0.288	0.063	**0.374**	0.247
Pgo	**0.346**	-0.263	0.122	0.244
Ngr	**0.373**	0.004	**0.39**	0.138
Pgr	**0.372**	-0.064	**0.359**	0.22
Pcq	0.17	**-0.569**	-0.167	**0.543**
Fanes	**-0.37**	-0.074	**-0.301**	**0.411**
TR	0.278	**0.435**	**0.406**	0.04
IR	**0.381**	0.103	**0.392**	-0.112

Bold values are significant for axis formation (≥ 0.3).

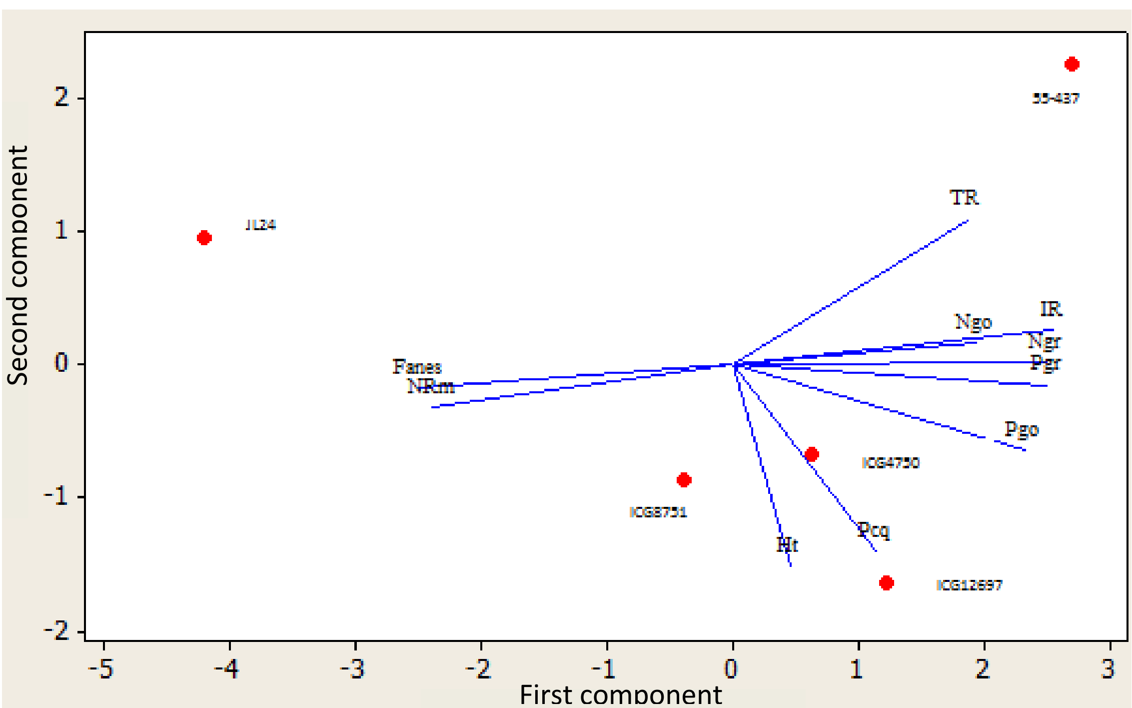

Figure 5. Double proportion diagram for plants under well watered conditions (T0).

In well-watered conditions, significant negative correlations were observed between the vegetative parameters (aerial biomass, Ht, NRm) with the yield parameters (Ngo, Pgo, Ngr, Pgr, IR). This means that when the water is not limited, the genotypes develop more aerial biomass. But there is an inter-genotypes difference. According to Gigih et al. (2018), there is a genotypic difference in plant grown under the same environmental conditions. ICG1269 has a very high height and empty pod weight and a relatively low pod

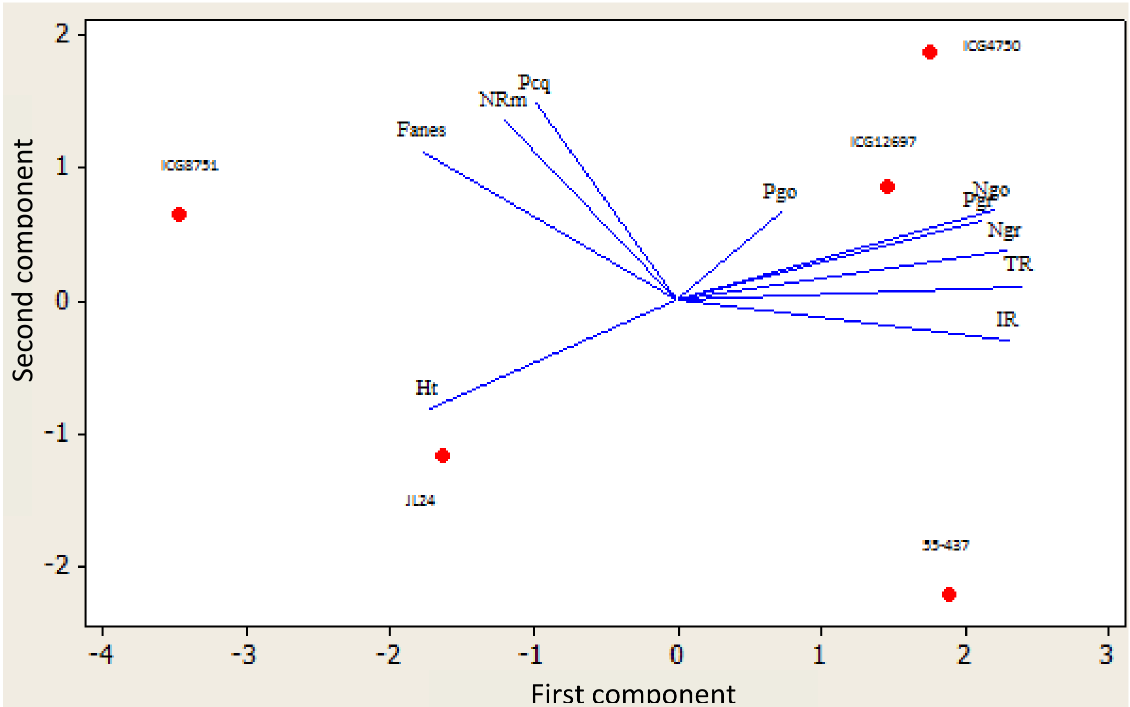

Figure 6. Double proportion diagram for plants under water deficit (T1).

filling rate unlike 55-437 and ICG4750. This means that at ICG12697, the pod filling time is relatively longer compared to 55-437 and ICG4750.

Nevertheless, when water is limited, plants slow down their growth by reducing the biomass in favor of pod filling. According to Gigih et al. (2018) under normal condition, groundnut plants are more focused on pod propagation to encourage more pods production. Under conditions that causes less pod formation, the plant focuses on seed enlargement. All genotype increase the number (NRm) of branches but the biomass and yield parameters at harvest reduced, due to leaves lost. These results corroborate with those of Mukhtar et al. (2014) who showed that the yield component of groundnut where affected by time and intensity of defoliation. Zhang and Uwe (2017) revealed that drought stress that occurs during plant growth will affect the plant growth. It may decrease plant yield during harvest. JL24 and ICG8751 produced fewer seeds compared to the other genotypes (55-437, ICG4750 and ICG12697). This low productivity is due to their strong vegetative growth, which would have accelerated the use of water and the decrease in soil water reserves, leading to more severe water stress in its latter.

Conclusion

The study of the water deficit on the agro-morphological responses of the genotypes studied made it possible to identify the best performers according to the objectives and production conditions. Indeed, for an objective of seed production in rain-fed culture in Niger, the genotypes ICG4750 and 55-437 can be proposed because of their high productivity under water deficit conditions and well-watered. JL24 can be proposed for irrigated crops, in particular for the production of biomass and for better integration of livestock farming in a context where fodder resources are becoming increasingly scarce.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Clavel D, Bradat P, Khalfaoui JL, Drame NK, Diop ND, Diouf O, Zuly-Fodil Y (2007). Adaptation à la sécheresse et création variétale : le cas de l'arachide en zone sahélienne. Deuxième partie: une approche pluridisciplinaire pour la création variétale. Oléagineux, Corps Gras, Lipides 14(3):293-308.

Dahanayake N, Ranawake AL, Senadhipathy DD (2015). Effects of water stress on the growth and reproduction black gram (*vigna mungo* L.). Tropical Agricultural Research and Extension 17(1):45-48.

Duncan W, McCloud D, MacGraw R, Boote K (1978). Physiological aspects of peanut yield improvement. Crop Science 18(6):1015-1020.

Bennett E, Jeremy A, Roberts, Carol W (2012). Manipulating resource allocation in plants. Journal of Experimental Botany 63(9):3391–3400.

FAO (2014). The state of world fisheries and aquaculture 2014, Food and Agriculture Organization of the United Nations (FAO), Rome 223 pp.

Gigih IP, Ropolia, Anaggaeni (2018). Selection of Bangka local groundnut (*Arachis hypogaea* L.) germplasm tolerant to drought stress. Advances in Engineering Research 167:183-187. https://dx.doi.org/10.2991/icoma-18.2019.39

Halilou O, Hamidou F, Boulama KT, Saadou M, Vincent V (2015). Water use, transpiration efficiency and yield in cowpea (*Vigna unguiculata* Walp L.) and peanut (*Arachis hypogaea*) across water regimes. Crop and Pasture Science 66:715-728.

Halilou O (2016). Effet de la sécheresse sur la reproduction et réponses physiologiques de l'arachide (*Arachis hypogaea* L.) et le niébé (*Vigna unguiculata* L. Walp.). Thèse doctorat, Université de Niamey 206 p.

Himeno K, Inoue N, Kurauchi N, Ito O, Nishikawa Y, Kobayashi Y (2009). Les céréales au Niger. *Accent sur le mil et le sorgho.* JAICAF (Association pour la Collaboration Internationale en matière d'Agriculture et de Forets du Japon). P. 109.

Madhusudhan KV, Giridarakumar S, Ranganayakulu GS, Reddy PC, Sudhakar C (2002). Effect of water stress on some physiological responses in two groundnut (*Arachis hypogaea* L.) cultivars with contrasting drought tolerance. Journal of Plant Biology 29:199-202.

Mukhtar AA, Falaki AM, Ahmad A, Jaliya MM, Abdoulkarim B(2014). Response of groundnut (*Arachis hypogaea* L.) varieties to varying defoliation intensities. Proceedings of the 4th ISOFAR Scientific Conference pp. 921-924.

RECA (2015). Résultats définitifs de la champagne agricole d'hivernage 2014 et perspectives alimentaires 2014-2015. P. 32.

Sharma PS, Sivakumar MVK (1991). Penetrometer soil resistance, pod number and yield of peanuts as influenced by drought stress. Indian Journal of Plant Physiology 34:147-152.

Vurayai R, Emongor V, Moseki B (2011). Effect of water stress imposed at different growth and developpement stages on morphological traits and yield of bambara groundnuts (*vigna subterranean* L. Verde). American Journal of Plant Physiology 6(1):17-27.

Zhang B (2015). Micro.RNA: a new target for improving plant tolerant to abiotic stress. Journal of Experimental Botany 66:1749-1761.

Zhang H, Uwe S (2017). Differences and commodities of plant responses to single and combined stress. The Plant Journal 90:839-855.

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Phytophthora seedling blight disease of cacao and its control measures

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Symptoms of *Phytophthora* seedling blight disease of cacao were found on 3 weeks old cacao seedlings at the nursery of Seed Production Division of Ghana Cocoa Board in New Tafo-Akim during routine surveys of cacao diseases in the Eastern Region of Ghana. Survey of seedlings in the nursery was conducted to determine disease incidence (DI). Heat sterilization of soil and drenching with Ridomil Gold plus 66 WP (6% metalaxyl-M and 60% copper (I) oxide) and Champion (77% cupric hydroxide) fungicides were evaluated for disease management. Symptoms of the disease started as vertical brown lesions above the cotyledon toward the top of cacao seedlings, causing the stems and leaves to wilt. Out of a total of 135,000 inspected cacao seedlings at the nursery, 2,525 (DI = 1.8%) seedlings were infected within one week of disease identification. Isolations from samples of infected tissues yielded *Phytophthora palmivora.* The fungicides were effective in disease control but heat sterilization of nursery soil before sowing cocoa beans was found to be the ideal control method.

Key words: Seedling blight, cacao, *Phytophthora palmivora,* fungicides.

INTRODUCTION

Globally, the genus *Phytophthora* causes more losses than any other disease of cacao (Bowers et al., 2001; Ploetz, 2016). Losses due to *Phytophthora* pod rot was estimated at 700,000 metric tons in 2012 (Ploetz, 2016). *Phytophthora* causes seedling blight (Guest, 2007) and the disease can cause 100% loss if not controlled (Peter and Chandramohanan, 2014). Seedling blight caused by *P. palmivora* results in wilting of stems and leaves, defoliation and eventual death within seven days after infection (Nur'Aini et al., 2016).

The survival of *P. palmivora* as mycelia and chlamydospores (thick-walled resistant spores) in the soil (Gregory and Maddison, 1981) makes it difficult to control and it is usually transported through partially infected seedlings or contaminated soil to newly established fields or increase inoculum population in already infected fields during transplanting of cacao seedlings. Combination of row cover and 0.2% copper oxide protects seedlings from *P. palmivora* attack (Nur'Aini et al., 2016). *Phytophthora* seedling blight is managed through integrated approach involving the use of phytosanitary practices, biocontrol agents and fungicides (Peter and Chandramohanan,

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2014).

In Ghana, cacao is usually cultivated using seedlings. To encourage the planting of improved planting materials for the sustainability of Ghana's cocoa production, the Seed Production Division (SPD) and Cocoa Health and Extension Division (CHED) of Ghana Cocoa Board (COCOBOD) collaborate to raise and distribute hybrid seedlings at no cost to farmers across the cocoa growing regions of the country for planting every year. The seedlings are raised in polythene bags filled with top soil under shade and watered periodically for 3 to 6 months before transplanting to the field (Opoku-Ameyaw et al., 2010). Survival of seedlings at the nurseries is threatened, among other factors, by a host of diseases including *Phytophthora* blight.

Phytophthora seedling blight is a sporadic disease but causes great loss in Ghana due to little knowledge on disease identification, cause and management among workers. The goal of this study was to make it easy for cocoa extension agents, workers and farmers to identify and manage *Phytophthora* seedling blight of cacao. The specific objectives were to describe the symptoms and confirm the pathogen of *Phytophthora* seedling blight identified at the nursery of Seed Production Division of Ghana Cocoa Board in New Tafo-Akim during routine surveys of cacao diseases in the Eastern Region of Ghana. It was also to evaluate heat-treated soil and fungicides for disease management.

MATERIALS AND METHODS

Symptomatic characterization, disease incidence and sampling

Symptoms resembling *Phytophthora* seedling blight disease of cacao were found on 3 weeks old cacao seedlings raised in black polythene bags under shade with a watering regime of 3 times per week at the nursery of Seed Production Division of Ghana Cocoa Board in New Tafo-Akim (06°22' N and 00° 36' W) in the East Akim district of the Eastern Region of Ghana. Each seedling was visually inspected for the symptoms. Disease incidence was expressed as a percentage of seedling infection. Infected stems and leaves of randomly selected cacao seedlings were collected in sterile polythene bags and taken to the laboratory for isolation of *P. palmivora.*

Isolation and characterization of *P. palmivora*

P. palmivora was isolated from infected stems and leaves following the procedure of Agrios (2005). Pieces of 4 mm from advancing margins of infected stems and leaves were surface sterilized in 70% ethanol for 60s and blot-dried between sterile filter papers. With a pair of sterile forceps, the pieces of infected tissues were placed on water agar and incubated in the dark at 28°C. After 48 h of incubation, tips of emerging fungal colonies were aseptically transferred onto V8 juice agar medium (200 ml V8 juice, 800 ml sterile distilled water, 20 g agar and 2.5 gl^{-1} $CaCO_3$) amended with 10 mgl^{-1} pimaricin, 100 mgl^{-1} vancomycin and 10 mgl^{-1} pentachloronitrobenzene to inhibit bacterial growth. Colony morphology of a 5-day old pure culture was recorded as growth pattern. For sporangial form and dimensions, mycelia mat from a 7-day old culture under continuous light at 28°C was placed on glass slide and stained with lactophenol blue. Description of sporangial shape, measurement of pedicel length, length (l) and breadth (b) of 50 caducous sporangia from each of 3 replicated plates were carried out under Leica CME compound microscope at ×400 magnification and l/b ratio calculated.

Pathogenicity test

Apparently healthy 3 to 4 months old green hybrid cacao pods were centrally inoculated with 10 mm disc plugs taken from the periphery of 3-day old pure culture of *P. palmivora*. The pods were placed on foam soaked with 500 ml sterile distilled water in plastic tray (70 cm × 60 cm × 15 cm) and incubated at 28°C for 7 days. Ten (10) cacao cores of 10 mm in diameter taken at 7 mm depth with sterile cork borer from *P. palmivora* infected cacao pods were completely buried in each of 10 bags of heat-treated soil (autoclaved at 121°C for 15 min) and watered (100 ml/bag) at 2 days interval for 2 weeks prior to the sowing of hybrid cocoa beans. Ten (10) bags of heat-treated soil without *P. palmivora* infected cacao cores were sowed with hybrid cocoa beans and included as control. The beans were watered (100 ml/bag) at 2 days interval. There were ten (10) replicated bags per treatment. The treatments were labeled and arranged in a completely randomized block design on raised platform in the greenhouse with alternating day and night at maximum and minimum temperature of 33°C and 21°C (CRIG Meteorological data) respectively. The number of infected cacao seedlings in each treatment were recorded and expressed as percentage of infection. Infected stems were sampled and taken to the laboratory for re-isolation of *P. palmivora*.

Baiting of *P. palmivora* from soil

Apparently sterile soil without cacao seedlings and *P. palmivora*-contaminated soil with infected cacao seedlings were collected from the nursery, where the disease outbreak occurred, for the test. Cacao cores of 10 mm in diameter taken at 7 mm depth with sterile cork borer from apparently healthy 3 to 4 months old green hybrid cacao pods were surface sterilized in 70% ethanol for 60s, rinsed in three changes of sterile distilled water and air dried on sterile filter papers under laminar air flow. Fifteen cacao cores were completely buried in each soil type and moistened with 10 ml of sterile distilled water. There were four replicated Petri dishes for each soil type containing 100 g of soil sampled from 5 nursery bags (20 g of soil from each bag). The plates were placed in plastic trays lined with wet foams and incubated at room temperature for 7 days. Cacao cores from the four plates were pooled together for each soil type and washed in four changes of sterile distilled water. They were air dried on sterile filter papers under laminar air flow. The number of uninfected, infected and disintegrated cacao cores were counted and expressed as percentage of infection.

Recovery of *P. palmivora* from infected cacao cores

P. palmivora infection of cacao cores was confirmed through its recovery on V8 juice agar medium amended with 10 mgl^{-1} pimaricin, 100 mgl^{-1} vancomycin and 10 mgl^{-1} pentachloronitrobenzene to inhibit bacterial growth. Tissue bits of approximately 0.4 mm^2 were excised from uninfected, infected and disintegrated cacao cores, surface sterilized in 70% ethanol for 60 s and rinsed in three changes of sterile distilled water. There were three replicated plates containing 5 tissue bits for each category of cacao core and incubated under light at 28°C for 3 days. Growth pattern of *P. palmivora* was recorded. Sporangial shape and dimensions were used to characterize the pathogen.

Disease control in a greenhouse study

In an attempt to prevent further spread of the disease, cacao seedlings showing symptoms of the disease including the polythene bags containing apparently *P. palmivora*-contaminated soils were destroyed by burning the seedlings and exposing the soil to sun. In a greenhouse experiment, each of ten (10) bags of apparently *P. palmivora*-contaminated soil collected from the nursery, where the disease outbreak occurred, were sowed with hybrid cocoa beans (1 per bag) and drenched with 100 ml/bag of either 50 g/15 L water of Ridomil Gold plus 66 WP (6% metalaxyl-M and 60% copper(I) oxide) or 100 g/15 L water of Champion (77% cupric hydroxide). In another treatment, ten (10) bags of apparently *P. palmivora*-contaminated soil were heat treated by autoclaving at 121°C for 15 minutes, allowed to cool and sowed with hybrid cocoa beans. Ten (10) bags of untreated apparently *P. palmivora*-contaminated soil sowed with hybrid cocoa beans were included as control. There were ten (10) bags/replicates per treatment and arranged in a completely randomized block design on raised platform in the greenhouse with alternating day and night at 33 and 21°C (CRIG Meteorological station) respectively. The beans were watered at 2 days interval. Number of seeds that germinated and the number of seedlings that showed symptoms of blight in each treatment were recorded and expressed as percentage of germination and infection respectively. Number of days to germination of cocoa beans in each treatment was recorded. Heights (cm) of cacao seedlings above cotyledon in the various treatments were measured with a ruler. The number of leaves was counted and girths (cm) at 2 cm above cotyledon were measured with a Vernier caliper at 8 weeks after sowing.

Data analysis

To determine the influence of treatments on height, girth and number of leaves on cacao seedlings, recorded data were statistically analyzed for significance using Analysis of Variance (ANOVA) in GenStat 11^{th} Edition statistical software (VSN International Limited). The level of significance was taken at 5% probability. Means were separated whenever the *F* test values were significant ($p<0.05$) using Duncan's Multiple Range Test.

RESULTS AND DISCUSSION

Disease incidence and characterization of disease symptoms

Symptoms of seedling blight disease were found on 3 weeks old cacao seedlings. Out of a total of 135,000 inspected cacao seedlings at the nursery, 2,525 seedlings were infected, representing 1.8% of disease incidence within one week of disease identification. This poses a threat to cacao cultivation in Ghana since almost half of the seedlings may be lost to the disease before the recommended transplanting age of 6 months. Symptoms of the disease started as a vertical brown lesion above the cotyledon (Figure 1a) or at the cotyledon (Figure 1b) toward the top of the cacao seedling. The lesion darkened as it spread toward the top causing the stem and leaves to wilt (Figure 1c). The stems weakened and bent down (Figure 1d). The symptom appeared as blight on leaves (Figure 1e). Defoliation (Figure 1f) and eventual death (Figure 1g) of cacao seedlings occurred at the later stage of infection. However, roots were not infected (Figure 1h). *Phytophthora* seedling blight disease of cacao is a major constraint to cacao seedling production and disease incidence decreases with increase in age of seedlings (Peter and Chandramohanan, 2014). Seedlings up to 4 months old are susceptible to blight (Lim, 1980) but escape the disease when they are 5 to 6 months old (Peter and Chandramohanan, 2014). Symptoms of the disease were similar to that reported by Nur'Aini et al. (2016).

Characterization of *P. palmivora*

Pure culture of the isolate produced a stellate-striate colony pattern with no aerial mycelium. Elliptical to ovoid, papillated and caducous sporangia with short pedicel ranging from 1.3 µm to 4.8 µm were observed. Sporangial length (*l*) and breadth (*b*) ranged from 35.0 µm to 57.5 µm (average 45.3 µm) and 30.0 µm to 40 µm (average 34.5 µm) respectively with an average *l*/*b* ratio of 1.3 µm. Numerous chlamydospores were observed with an average diameter of 36.1 µm. Identification of *P. palmivora* as the cause of seedling blight confirms earlier reports (Peter and Chandramohanan, 2014; Nur'Aini et al., 2016). Colony morphology of the *P. palmivora* isolate was in agreement with previous observations (Opoku 2004). Sporangial shape and dimensions were within the ranges reported for *P. palmivora* (Erwin and Ribeiro, 1996; Drenth and Sendall, 2001). All major organs of cacao, including pods, stems, leaves and roots can be attacked by *P. palmivora* (Firman, 1974). *P. palmivora* is widely distributed (Bong et al., 2000; Thanh et al., 2004) and less virulent than *Phytophthora megakarya*, which is restricted to West Africa (Opoku et al., 2000). However, due to its presence in all cocoa producing areas, *P. palmivora* causes the greatest yield loss of cocoa (Ploetz, 2016). The pathogen requires wet conditions and high humidity for infection (Gregory and Maddison, 1981). Seedling infections were reported in June during the rainy season in India (Peter and Chandra, 2011, 2014). On the contrary, seedling blight was identified in the dry season of December at New Tafo-Akim in the Eastern Region of Ghana. Generally, the prevalence of *Phytophthora*

Figure 1. Symptoms of *Phytophthora* seedling blight disease showing brown lesion on the stem above the cotyledon toward the top (a), brown lesion from the cotyledon toward the top (b), wilting of stem and leaves (c), bending of seedling stem at the brown lesion toward the top (d), leaf blight (e), defoliation (f) die-back (g) and uninfected roots (h).

diseases of cacao is less in Ghana during the dry season (November to May annually). However, *Phytophthora* infections occur in West Java in the absence of rain as the humidity of nearly 100% that occur for a few hours during the night provide enough free water to initiate infection (Purwantara and Pawirosoemardjo, 1990; Purwantara, 2003). Perhaps, the cacao seedlings were infected in the dry season of December following continuous watering of seedlings under shade which enhanced germination and spread of propagules of *P. palmivora*.

Pathogenicity test

P. palmivora infected cacao seedlings were characterized by vertical brown lesions on stems (Figure 2) above soil level toward the cotyledon 11 days after germination. About 70% of infected cacao seedlings wilted and died. However, seedlings in sterilized soil remained healthy 44 days after germination. *P. palmivora* was successfully re-isolated from infected stems. Infections of stems above soil level toward the cotyledon contradicted field observations where the lesions appeared above the cotyledon or at the cotyledon toward the top of the cacao seedling. However, observed symptoms in both instances were caused by *P. palmivora*. It is unclear how the pathogen infected stems above cotyledon without causing root rots of seedlings at the nursery. It is suggested that water splash of contaminated soil might have introduced the pathogen to infect the aerial part of the stem above the cotyledon. The epicotyls or cotyledons might have also been infected by the pathogen before emerging from the soil.

Figure 2. Vertical brown lesion on the stem above soil level toward the cotyledon of *P. palmivora* infected cacao seedling.

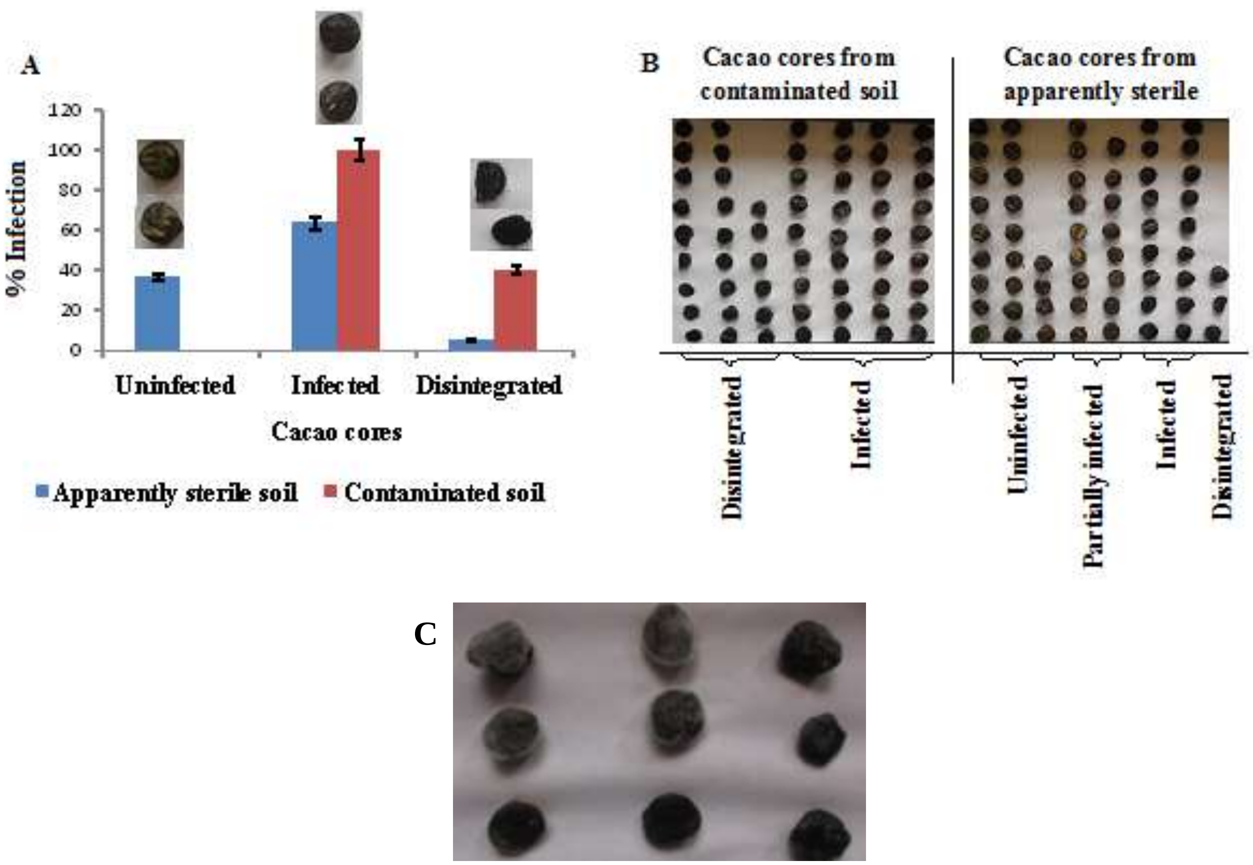

Figure 3. Graphical presentation of percentage of uninfected, infected and disintegrated cacao cores in apparently sterile nursery soil and *P. palmivora*-contaminated soils (A), Photographic presentation of uninfected, infected and disintegrated cacao cores in apparently sterile nursery soil and *P. palmivora*-contaminated soils (B), Infected cacao cores covered with bloom mass of *P. palmivora* after 24 h of incubation (C).

Table 1. Percent germination and infection of cacao seedlings in contaminated, sterilized, Champion and Ridomil Gold plus 66 WP treated soils.

Treatment	Cacao seedlings	
	% Germination	% Infection
Contaminated soil	70	40
Sterilized soil	90	0
Champion	90	10
Ridomil Gold plus 66 WP	60	0

Baiting of *P. palmivora* from soil

Infected cacao cores were characterized by brown lesions typical of *P. palmivora* infection (Opoku, 2004). All cacao cores in apparently *P. palmivora*-contaminated soil were infected, resulting in tissue disintegration of 40%. However, 36.7% of cacao cores which were not infected in apparently sterile soil remained green (Figure 3A). Out of 38 infected cacao cores in apparently sterile soil, only 3 were disintegrated (Figure 3B). Infected cacao cores were covered with bloom mass of hyphae (Figure 3C) after 24 h of incubation. Successful baiting of *P. palmivora* from the soil in 7 days demonstrates survival of the pathogen in soil. Baiting technique has been used to isolate *P. palmivora* from soil in 4 days (Okaisabor, 1971). It provides a quick and simple way of detecting the presence of the pathogen in the soil. Reports suggest that *P. palmivora* can survive in the soil up to 10 months (Enriquez and Zentmyer, 1980). This is an indication that propagules of the pathogen can be found in the soil all year round. In the dry season, the pathogen survives as chlamydospores (thick-walled resistant spores) or mycelia (Gregory and Maddison, 1981), making the soil a reservoir of the pathogen's inoculum. Under favourable conditions, the propagules germinate to infect roots of cacao seedlings and subsequently infect aerial parts of the seedling through water splashes of contaminated soil during watering. Inoculum size of the pathogen increase in the soil after root infections and this was evidenced in the infection of all cacao cores in the apparently *P. palmivora*-contaminated soil. Due to increase in population size of the inoculum in the contaminated soil, cacao cores were probably infected in a few days leading to the disintegration of 40% after 7 days of incubation.

Recovery of *P. palmivora*

P. palmivora was successfully recovered from infected and disintegrated cacao cores. Pure cultures of isolated *P. palmivora* produced stellate-striate colony patterns on V8 juice agar medium. However, no growth was observed on V8 juice agar plates inoculated with tissue bits excised from uninfected cacao cores 3 days after incubation.

Disease control in a greenhouse

In Ridomil Gold plus 66 WP treated soils, 60% of cocoa beans germinated and none of the seedlings was infected. Similarly, all seedlings in sterilized soils remained healthy 43 days after germination. On the contrary, 40% of seedlings were infected in contaminated soil (Table 1). There were no significant (p>0.05) differences in the number of days for germination of cocoa beans, heights and number of leaves on cacao seedlings in the various treatments. However, girths of cacao seedlings in sterilized soil were significantly (p=0.027) bigger than those in the other treatments (Table 2). As a soil-borne pathogen, control of *Phytophthora* is a challenge (Erwin et al., 1983). Burning of infected cacao seedlings, including the polythene bags containing apparently *P. palmivora*-contaminated soils, and exposing the soil to sun prevented further spread of the disease at the nursery. Opoku et al. (2007) reported that fungicide application combined with crop sanitation practices were effective in the management of *Phytophthora* on cacao. In this study, drenching of soil with Ridomil Gold plus 66 WP (6% metalaxyl-M and 60% copper (I) oxide) and Champion (77% cupric hydroxide) were effective in the control of the disease even though 10% of the seedlings were infected in Champion-treated soils. Despite the effectiveness of Ridomil Gold plus 66 WP in the control of the disease, only 60% of the cocoa beans germinated after 13 days of sowing. Low germination of cocoa beans in Ridomil Gold plus 66 WP treated soil could be due to the fungicide since negative influence of long-term use of pesticides on microbial growth and activity, leading to reduced soil fertility and productivity has been reported (Wang et al., 2006). Braun and Supkoff (1994) reported that soil treatment by steaming at 80 – 100°C for half an hour effectively controls most soil-borne pathogens. In this study, *P. palmivora* was effectively controlled as all the germinated cocoa beans (90% germination) in the sterilized or heat-

Table 2. Number of days for germination of cocoa beans, girth, height and number of leaves on cacao seedlings in contaminated, sterilized, Champion and Ridomil Gold treated soils.

Treatment	Number of days for germination of cocoa beans	Girth (cm)	Height (cm)	Number of leaves
Ridomil Gold Plus 66 WP	12	30.6[b]	24.3	10
Champion	13	32.0[b]	21.6	11
Sterilized soil	13	36.5[a]	23.8	11
Contaminated soil	12	30.5[b]	21.9	10
lsd	Ns	4.2	ns	ns[#]

Mean followed by a different letter is significantly different at p=0.027. ns[#] = Values are not significantly different from each other at p=0.05.

treated soil survived for 43 days without infection, making it the ideal control method.

Conclusions

The importance of seedling blight disease of cacao is highlighted in this study. Symptoms of the disease have been described in this paper to make it easy for cocoa extension agents, workers and farmers to identify it. Heat sterilization of soil and the use of fungicides for disease management have been demonstrated. However, sterilization of nursery soils is recommended to kill inoculum of *P. palmivora* and any other pathogen that may infect cacao seedlings.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Agrios GN (2005). Plant pathology (5th edition). USA. Elsevier Academic Press, pp. 398-402.

Bong CL, Chong TC, Lim KL, Lim GT (2000). Integrated crop management of cocoa clonal plantings for higher productivity and efficiency of crop protection in Sabah, Malaysia. In. Bong CL, Lee CH, Shari FS (Eds.), Proceedings of 3rd INCOPED International Seminar. Kinabalu, Malaysia, pp. 123-142.

Bowers JH, Bailey BA, Hebbar PK, Sanogo S, Lumsden RD (2001). The impact of plant diseases on world chocolate production. Online. Plant Health Progress. doi:10.1094/PHP-2001 0709-01-RV.

Braun AL, Supkoff DM (1994). Options to methyl bromide for the control of soil-borne diseases and pests in California with reference to the Netherlands. California Department of Pesticide Regulation. Sacramento, CA: Environmental Protection Agency, Environmental Monitoring and Pest Management Branch.

Drenth A, Sendall B (2001). Practical guide to the detection and identification of *Phytophthora* (Version 1.0). Brisbane, Australia. CRC for Tropical Plant Protection, pp. 1-41.

Enriquez GA, Zentmyer GA (1980). Proceedings of the meeting of the American Regional Group of *Phytophthora palmivora* on cocoa. Turrialba, Costa Rica. CATIE. Technical Report 126:40

Erwin DC, Bartnicki-Garcia S, Tsao PH (1983). *Phytophthora*: Its biology, taxonomy, ecology, and pathology. St. Paul, MN, USA. APS Press 392p.

Erwin DC, Ribeiro OK (1996). *Phytophthora* diseases worldwide. St. Paul, MN, USA. APS Press.

Firman ID (1974). Cocoa canker. In. Gregory PH (Ed.). Phytophthora Disease of Cocoa. Longman, London. pp. 131-140.

Gregory PH, Maddison AC (1981). Epidemiology of *Phytophthora* on Cocoa in Nigeria. Phytopathological Paper 25:23-26.

Guest D (2007). Black pod: Diverse pathogens with a global impact on cocoa yield. Phytopathology 97:1650-1653.

Lim TM (1980). A forecasting system for use in the chemical control of *Phytophthora* leaf fall on plantation rubber in Malaysia. Paper presented at workshop on *Phytophthora* diseases on tropical cultivated plants, Kerala, India.

Nur' Aini F, Santoso TI, Sudarsianto S (2016). Application of copper fungicide and row covers to control *Phytophthora* seedling blight disease on cocoa. Pelita Perkebunan 32(2):101-108.

Okaisabor EK (1971). The mechanism of initiation of *Phytophthora* pod rot epiphytotoics. Proceedings of the 3rd International Cocoa Research Conference. Accra, Ghana. pp. 398-404.

Opoku IY, Appiah AA, Akrofi AY, Owusu GK (2000). *Phytophthora megakarya*: a potential threat to the cocoa industry in Ghana. Ghana Journal of Agricultural Science 33:237-248.

Opoku IY, Assuah MK, Aneani F (2007). Management of black pod disease of cocoa with reduced number of fungicide application and crop sanitation. African Journal of Agricultural Research 2(2):601-604.

Opoku IY (2004). Field and laboratory characterization of *Phytophthora palmivora* and *Phytophthora megakarya* in Ghana. Technical Bulletin No. 15. Cocoa Research Institute of Ghana, Akim-Tafo 24 p.

Opoku-Ameyaw K, Baah F, Gyedu-Akoto E, Anchirinah V, Dzahini-Obiatey HK, Cudjoe AR, Aquaye S, Opoku SY (2010). Cocoa Manual: A source book on cocoa production (1st Edition). Cocoa Research Institute of Ghana, Akim-Tafo, pp. 14-19.

Peter PK, Chandramohanan R (2011). Occurrence and distribution of cocoa (*Theobroma cocoa*, L.) diseases in India. Journal of Research ANGRAU 39:44-50.

Peter PK, Chandramohanan R (2014). Management of cocoa seedling blight caused by *Phytophthora palmivora*. Plant Archives 14(1):461-463.

Ploetz R (2016). The impact of diseases on cacao production: a global overview. In. Bailey BA, Meinhardt LW (Eds.). Cacao diseases. New

York, USA: Springer International Publishing, pp. 33-59.
Purwantara A (2003). Epidemiology and control of *Phytophthora* diseases of cocoa in Java, Indonesia. Proceedings of 8th International Congress of Plant Pathology. Christ church, New Zealand. Sydney, Australia. Horticulture Australia 353 p.
Purwantara A, Pawirosoemardjo S (1990). Fluktuasi intensitas penyakit *Phytophthora* pada buah kakao di daerah basah. Menara Perkebunan 58:44-50.
Thanh DVT, Vien NV, Drenth A (2004). *Phytophthora* diseases in Vietnam. In. Drenth A, Guest DI (Eds.). Diversity and management of *Phytophthora* in Southeast Asia. ACIAR, Canberra, Australia, pp. 83-89.
Wang MC, Gong M, Zang HB, Hua XM, Yao J, Pang JY, Yang YH (2006). Effect of methamidophos and urea application on microbial communities in soils as determined by microbial biomass and community level physiological profiles. Journal of Environmental Science and Health, Part B: Pesticides, Food Contaminants, and Agricultural Wastes 41(4):399-413.

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Selenium in Caprine feeding, its transference to the milk and relation with other minerals

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The experiment was performed in the campus of the State University of Alagoas, in Santana do Ipanema, in the semi-arid region of Northeast Brazil. This work is aimed at studying the transferences of Se from animals' food to milk in various levels of offers and its correlations with Ca, P, Mg and soluble Fe. The work also analyzed the somatic cells counts, total bacteria count and chemical composition of milk: Fat, total soluble solids, lactose and proteins. Eight Anglo Nubians animals with a live weight of 60 kg with fifteen days of second lactation were stabled in individual stalls and confined in a completely randomized design, DIC. The animals were fed with 2 kg.day^{-1} of cactus pear and 0.5 to 1.0 kg.day^{-1} Tifton hay (*Cynodon* spp.); concentrated and formed with 333.3 g.day^{-1} corn and 166.67 g.day^{-1} soy. The mineral salt was dosed in three treatments using selenium (Se). The treatments were; Treatment 00: at 0.0 mg kg^{-1} of animal MS.day^{-1} serving as control group with no supply of mineral salt, and if provided, it came from their own diet; Treatment 01: at 0.1 mg MS.day^{-1} of animal.kg-^{1}; Treatment 02: at 0.45 mg of animal. kg^{-1} DM. day^{-1}; and Treatment 03: with 0.90 mg Se animal.kg^{-1} DM.day^{-1} ($p < 0.05$) using Tukey test.

Key words: Calcium, phosphorus, magnesium, soluble iron.

INTRODUCTION

The secretion of the milk itself begins right after the colostrum period and its composition varies in function of the goat's feeding, race, age, the volume of milk produced and the lactation phase. However, the several constituents of milk are produced in many different ways; the fat is synthesized in the alveoli, from the fatty acids contained in the blood, with fermentation occurrence in the rumen. The main fat is the acetic acid. A goat can produce 200 or even 250 g of fat per day.

Milk protein is synthesized in the alveoli as well, from the blood amino acids. The main milk proteins non-existent in the blood are casein and lactalbumin. Lactose is also produced in the alveoli from glucose. Minerals are present in the milk and on the blood in different proportions: milk has 10 to 15 times more calcium, potassium and phosphorus than blood and three times

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less chlorine. The vitamins pass the alveoli walls unaltered and vary in function of feeding and breeding method (Power and Dhamoon, 2019).

Goat milk is one of the main foods of many Brazilian children, mainly in the countryside of Brazil's semi-arid areas and is very rich in protein, lactose, vitamins and mineral salts and is of easy absorption (Leite et al., 2017). It works as tissue protector against oxidative stress, maintenance of the defenses against infections and conformation of the body's growth and development on the human body (Pizzino et al., 2017).

Selenium is an important antioxidant. There are two isoforms of glutathione peroxidase, one selenium dependent (Se/GPx) and one selenium independent (Se/GPx). The Se independent form is found at the cytosol and does not present great H_2O_2reduction capacity. The GPx presents bigger affinity to the hydrogen peroxide when it finds itself in big concentrations at the cytosol (Ferreira and Matsubara, 1997).

Brazil food has low levels of Selenium, thus requiring daily complements to obtain the recommended levels (Silva et al., 2007). The products of vegetal origin present less Se concentration than the ones of animal origin. So milk is one of the products that provide a good quantity of selenium to supply human needs with minerals.

Ghany-Hefnawy and Tortora-Perez (2008) affirm that selenium (Se) is an essential mineral on the animal nutrition and it is associated with many animal production processes, such as species fertility and diseases prevention. The first enzyme where the active presence of selenium and its importance to avoiding the oxidative damage at the cellular membranes was demonstrated was the glutathione peroxidase (GSH-Px).

One of the main roles of selenium on the organism is to be antioxidant, this way it is always associated to the enzyme glutathione peroxidase. Besides acting on the detoxification of the hydrogen peroxide and other organic peroxides, the glutathione peroxidase acts on the maintenance of vital sulfhydryl groups in the reduced form, on the synthesis of hormones and on the metabolism of compounds strange to the organism as well; for example, aromatic compounds derived from plants and pesticides (Kurutas, 2016).

The Selenium (Se) is an example of an essential element becoming more and more insufficient in food crops as a result of intensive plant production in many countries. Se is an essential biological trace element. It is an essential constituent of several enzymes in which it is present in the form of the unusual amino acid selenocysteine (SeCys)(El-Ramady et al., 2015). Few researches show the Se concentration in cow and human milk and there is no thorough research on goat milk in Brazil.

To Hunter et al. (2005), selenium and vitamin E are interdependent, both needed for animals and both have metabolic functions on the organism, besides an antioxidant effect. In some cases, one can replace the other or even slow down the effects of vitamin E deficiency.

The Se shortage causes serious problems on the productive efficiency and animals' health, as the mortality of offspring with severe disabilities, as consequence of myocardial lesions. Between the anomalies studied and documented, they showed less gain of weight, less milk and wool production, low reproductive efficiency, with fertility reduction, the prolificacy and the animal quality (Ghany-Hefnawy and Tortora-Perez, 2010).This work aims to study the transference of Se from animal food to the milk in various levels of offers, as well as its correlations with Ca, P, Mg and soluble Fe. The somatic cells counts, total bacteria count and chemical composition of milk: Fat, Total Soluble Solids, Lactose and Proteins were also analyzed.

MATERIALS AND METHODS

The experiment was conducted on the Campus II of UNEAL in Santana do Ipanema, with 8 goats from the research project of APL Ovinocaprinocultura. This work observed the levels of Calcium, Phosphorus, Selenium and soluble Iron in the goat milk, using three treatments by adding sodium selenite, as well as the physical-chemical properties of milk in its most characteristic composition. The analysis of the minerals Se, Ca, P, Mg and Fe were realized on the labs of the company, Qualitex, located in Marechal Deodoro, AL, Brazil.

The analysis of the percentage of fat, protein, lactose and total solids; somatic cells count by Ml × 1000 and total bacteria count by mLx1000 were realized in the lab, PROGENE,of the Northeast Dairy Herds Management Program, from the Zootechnics Department, Federal Rural University of Pernambuco, Brazil.

Collection of milk samples from the dairy goats

Eight Anglo-Nubian animals with live weight of 60kg, with fifteen days from the second lactation, were stabled in individual stalls and confined in an entirely randomized lineation. They were fed with 2 kg.day^{-1} of forage palm, from 0.5 to 1.0 kg.day^{-1} of Tifton grass hay (*Cynodon* spp.) and concentrate formed by 333.3 g.day^{-1} of corn and 166.67 g.day^{-1} of soy.

The mineral salt was dosed on the three treatments of Selenium (Se) usage, this way the treatments were as follows: 0,0 mg of animal Se.kg^{-1} MS.day^{-1}to the control group – there was no supply of mineral salt, the Se provided came from the diet itself; the second with supply of 0.1 mg of animal Se.kg^{-1} MS.day^{-1}; and the third were provided with 0.45 mg of animal Se.kg^{-1} DM. day^{-1}, level 1 proposed; the forth with 0.90 mg of animal Se. kg^{-1} DM.day^{-1},level 2 proposed. The selenium (Se) was provided on the form of sodium selenite (Na_2SeO_3) powder, added to the mineral salt; the mix was supplied on the dosage of 40 g of animal mineral salt.day^{-1} mixed and added to the soy as well as corn concentrate and given at 16h00min (Pizzino et al., 2017). Mineral salt was provided with the composition of 15 mg.kg^{-1}; being proportionally provided with the dosage of the treatment, 01:0.1mg animal Se.kg^{-1}DM.day^{-1}.

Basic composition of the mineral salt Caprinofós - Tortuga© (2016)

The basic composition comprised Calcium Phosphate, Potassium Chloride, Calcium Carbonate, Vitamin E, Carbon-Amino- Zinc

Table 1. Composition of the mineral salt provided to the animals on the experiment by 01kg of the product, guarantee levels (Mineral salt to caprineCaprinofósTortuga© (2016).

Component	Quantity (unit)
Calcium	240 g
Phosphorus	71 g
Potassium	28.20g
Sulfur	20.00g
Magnesium	20.00g
Iron	2.500 mg
Zinc	1.700 mg
Manganese	1.350 mg
Fluorine (Max.)	710 mg
Copper	400 mg
Iodine	30 mg
Cobalt	30 mg
Selenium	15 mg
Chromium	10 mg
Vitamin A	135.000 UI
Vitamin D3	68.000 UI
Vitamin E	450 UI
P solubility in citric acid	2 a 95 %

Table 2. Daily needs for the maintenance of confined caprine with 60 kg of live weight (PV) and daily production of 0.5 kg of milk with 3.0% of fat.

Items	DM (kg)	PB (%)	EM (Mcal)	NDT (g)	Ca (g)	P (g)	Se ($mg.Kg^{-1}DM$)	Fe ($mg.kg^{-1}DM$)
Quantitiy	1.09	118	2.79	945	4	2.8	0.1	50

Source: Andriguetto (1983).

Phospho Chelate of Zinc, Copper, Selenium, Manganese and Chromium, Trans-chelated Micromineral Premix, Vehicle Q.S.,Magnesium Oxide, Vitamin D3, Vitamin A, Ventilated Sulfur (Sulfur Flower) (Table 1).

The stabled animals received the proposed diet, with the mineral salt in different treatments every fifteen days. The milk sample were collected and moved forward to the next treatment; that is, every treatment had fifteen days of adaptation to the diet, in order to collect the milk samples. Milk samples were collected at the end of every fifteen days in sterile polyethylene flask with 100mL volume, packed on thermal boxes in low temperatures and sent to PROGENE and Qualitex labs for analysis. The provided diet was calculated based on the indications of daily needs recommended by Andriguetto (1983),related to the contents of dry matter (MS) in Kg, crude protein (CP) in percentage (%), metabolizable energy (ME) in megacalories (Mcal), total digestible nutrients (TDN) in grams, Calcium (Ca) in grams, Phosphorus (P) in grams, Selenium (Se) in milligram by kilogram of dry matter ($mg.kg^{-1}DM.^{-1}$) and Iron (Fe) in milligram by kilogram of dry matter ($mg.kg^{-1}M.S.^{-1}$) (Table 2).

The daily diet provided by stabled animal by Day was composed of small forage palm (2 $kg.day^{-1}$), Tyfton hay (1 $kg.day^{-1}$), concentrate containing 152.5g of soy bean meal, 347.5g of powdered corn and 40g of mineral salt for caprine. The ingredients composition can be found on chart 5.3. The provided ingredients data were analyzed at the Qualitex lab, in Marechal Deodoro, Alagoas, by Andriguetto (1983) and National Research Council (2001).

The forage palm, *Nopanalea cochonilifera,* is most used locally, because this cactus adapts well to the semi-arid and today represents the second most cultivated culture on the state of Alagoas, after sugar cane. Also, it serves as roughage; utilized hay of the *Cynodon* type, the *Tyfton*gramineous, the most important gramineous in the hay production today on Brazil's northeast. The bromatological composition of the nutrients of the diet used in thestudy was based on the following analysis: dry matter (DM), crude protein (CP), ethereal extract (EE), crude fiber (CF), total digestible nutrients (TDN), metabolizable energy (ME), Calcium (Ca), Phosphorus (P), Selenium (Se) and soluble iron (Fe) (Table 3). Analyses were carried out on Qualitex labs, consistent with Studies by Andriguetto (1983) and National Research Council (2001).

The offered concentrate (0.5 kg) formed by 30.5% of soybean meal (*Glycine max* L.) as protein source and 69.5% of crushed corn grain (*Zea mays* L.) supplied daily through soy and milk of 68.60 and 31.40 g of crude protein respectively. This corresponds in metabolizable energy to 0.401 and 0.990 Mcal for soy and corn respectively. The roughage provided had as base the grass hay Tifton (*Cynodon* spp.) of 1$kg.day^{-1}$and small forage palm (*Nopalea cochonilifera*) of 2 $kg.day^{-1}$; supplying 102.50g and 10.0g of crude protein and 1.515 and 0.286 Mcal of metabolizable energy for hay and palm respectively (Table 4).

Table 3. Contents of dry matter (DM), crude protein (CB), ethereal extract (EE), crude fiber (CF), total digestible nutrients (TDN), metabolizable energy (ME), calcium (Ca), phosphorus (P), Selenium (Se) and soluble iron (Fe) of the ingredients used on the diet of the animals stabled for the study.

Items	Tyfton 68 hay	Forage palm	Soybean meal	Powdered corn
Dry matter (MS) %*	89.71	10.53	87.16	87.19
Crude protein (PB) %**	10.25	5.00	45.00	9.30
Ethereal extract(EE) %**	1.80	2.40	0.90	4.30
Crude fiber (FB) %**	28.30	8.00	6.00	2.00
Total digestable nutrients (NDT) %**	55.00	***66.16	73.00	80.00
Metabolizable energy (EM) Kcal/kg**	1.515.00	***1.430.00	2.639.00	2.846.00
Calcium (Ca) g/100g*	0.32	0.03	0.28	0.02
Phosphorus (P) g/100g*	2.14	0.065	1.48	0.37
Selenium (Se) g/100g*	0.001	0.001	0.001	0.001
Soluble iron (Fe) g/100g*	1.039	0.022	1.73	0.013

*Analysis realized on the Qualitex lab - Al. **(Andriguetto, 1983); *** (NRC, 2001).

Table 4. Contents of crude protein (CP) and metabolizable energy (ME), balance for the daily diet supplied to maintain 60 kg caprine and production of 0.5 kg of milk with 3% of fat.

	CP (g)	ME (Mcal)
Concentrate (soy + corn) (0.5 kg)		
Soy (30,5%)	68,60	0.401
Corn (69,5%)	31,40	0.990
Subtotal	**100.00**	**1.390**
Roughage (hay + small palm) (3.0 kg)		
Hay (1kg)	102,50	1.515
Palm (2kg)	10,00	0.286
Subtotal	**112.50**	**1.804**
Total	**212.50**	**3.191**

Values quoted by Andriguetto (1983).

Processing of the collected samples

At the PROGENE lab, the samples were analyzed using the method of flow cytometry to total bacteria count and somatic cells count and milk composition with infra-red Bentley – Infra Bentley 2000 Sumercont 300.The mineral contents were analyzed on the Qualitex lab in an atomic absorption device (using acetylene flame) coupled to a hydride generator. The calibration curve was prepared with a solution proper to atomic absorption. The analyses were realized according to a methodology quoted by Greenberg et al. (2012).Calcium SMEWW 3111 B; Selenium SMEWW 3113 B; Humidity Pl122; Phosphorus SMEWW 4500:Method of ascorbic acid; Iron SMEWW 4500 Method of phenanthroline

Data analysis

The data were analyzed statistically in its diverse correlations between Se and other items to analyze the mineral influence on the different dosages, utilizing the Origin program version 8 and analyzed by the Tukey test $p < 0.05$.

RESULTS AND DISCUSSION

The development of these studies brought possibilities of chemical evaluations in the comparison of influences of the many minerals studied and in the general composition of the analyzed goat milk.

The result of Se levels on the milk was always inferior to 0.001 mg.100 ml^{-1}. This way, all the results obtained on every milk analysis were 0.001 mg.100ml^{-1}. Similarly, the considered values of Se were 0.001 mg.100g^{-1}.

The results suggest that with mineral salt application, in general, there was an increase in the Ca levels on the milk, proving a direct transfer between the diet and the quantity of Ca provided on the milk. Effectively, the different levels of Se on the diet did not interfere with the Ca levels provided by the result of milk contents (Figure 1). The Ca levels were elevated from an average of 60 mg.100 ml^{-1} to results above 140 mg.100 ml^{-1}, more than

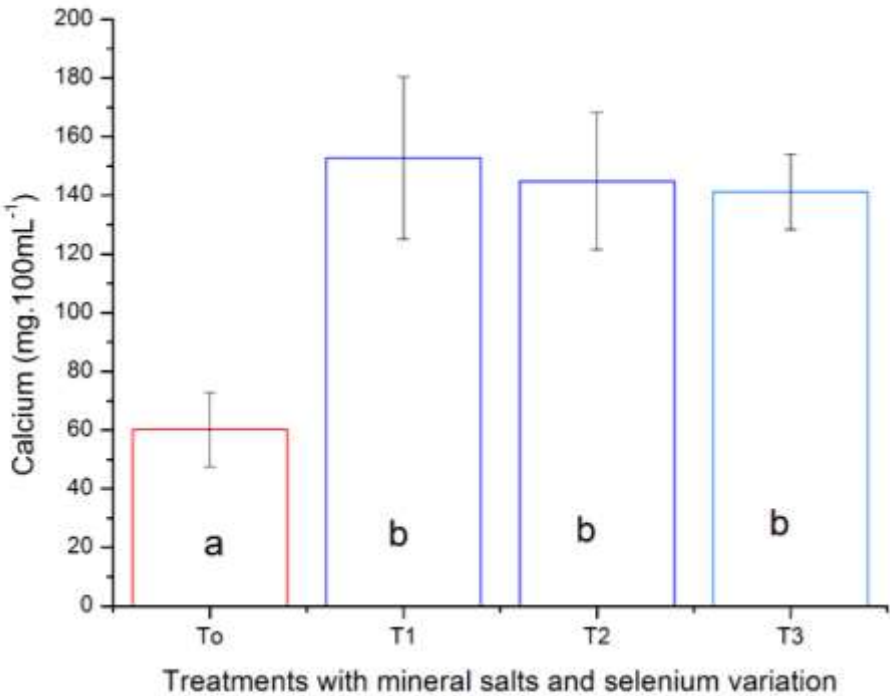

Figure 1. Calcium levels on goat milk with adding of mineral salt and three levels of selenium. The bars represent the average and the vertical lines the standard errors (n=32). Different letters represent significant difference related to the control group ($p<0.05$) by the Tukey test.

the double of Ca level of the control group; which is due basically to the simple supply of mineral salt on the caprine diet.

Nascimento et al. (2008) studied the effect of dietetic supplementation of Selenium (Se) on the production and quality of the caprine embryo: the control group received 25g of mineral salt that contained 30 mg of Selenium.Kg^{-1} and the experimental group received 25g of mineral salt that contained 90mg of Selenium.Kg^{-1}. The study showed that for the embryos of degree 1 (5.46 ± 1.37) and infeasible (2.68± 1.36) bigger on the control ($p<0.05$) and 3.98 ±1.48 of the embryos of group 2 is bigger on the treated ones ($p<0.05$). The authors concluded that there was no difference ($p>0.05$) between the initial selenium plasmatic levels between the two groups and that the mineral salt administration containing 30mg of selenium.kg^{-1} is enough to produce quality caprine embryos.

According to Khalili et al. (2020) there was no significant difference in haematological parameters before and after delivery in experimental and control groups. However, in the prepartum period, MCH tended to increase significantly in selenium methionine treatment ($p < 0.05$). The mean of rectum temperature in the treatment of selenium methionine was significantly lower than that of the control group ($p < 0.05$). On the other hand, the purulent vaginal content, retained placenta, and mastitis were lower in this group. The results of this experiment showed that feeding organic selenium supplementation in multiparous dairy cow's diet may improve their health and reproduction.

The study takes into consideration that the Phosphorus levels did not obtain the same behavior like Calcium. The control group presented results around 32 mg.$100ml^{-1}$ of P and lower results for treatments 1 and 2 with additional applications of selenium with an average of 22 mg.$100ml^{-1}$ of P. The treatment 3, however, presented a recovery and an overcoming of P value on the significant milk ($p<0.05$) to the increase of selenium on the lactating goat diet (Figure 2).

Brasil et al. (2000) in a study on goats under neutral thermal zone and under stress obtained Phosphorus93.36 mg/dL in the morning and in the afternoon an average of 88.92 mg/dL. Souza et al. (2013) cited thatunder the conditions of the present study, it is concluded that the goats were influenced by climatic factors, where the rainy period was more likely to cause thermal stress in the animals.

The results of soluble iron suggest, such as the calcium results, a raise on the values of treatments 1 to 3 (from 1.4 mg.$100ml^{-1}$ to 1.6 mg.100 ml^{-1}) in relation to the control group (0.5 mg.$100ml^{-1}$). However, it suggests, as well, that the variations of selenium offers on the diet did not interfere with the results, because the differences

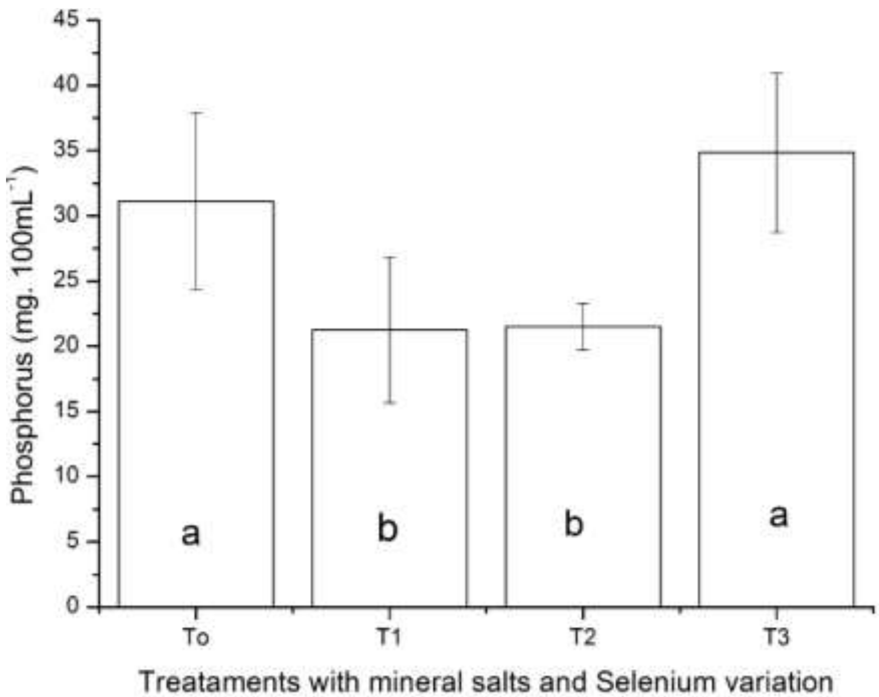

Figure 2. Phosphorus levels on goat milk with adding of mineral salt and three levels of selenium. The bars represent the averages and the vertical lines the standard errors (n=32). Different letters represent significant difference related to the control group (p<0.05) by the Tukey test.

between the treatments were not significant ($p > 0,05$) to the following treatments: Treatment 00, on the levels 0.0 mg of animal Se.kg^{-1} MS.day^{-1}; Treatment 01, with supply of 0.1 mg of animal Se.kg^{-1} MS.day^{-1} according to Andriguetto (1983) recommendations; and Treatment 03, with 0.90 mg animal Se.kg^{-1} DM.day^{-1}. Although significant ($p < 0.05$), Treatment 02 provided 0.45mg animal Se. kg^{-1} DM. day^{-1} (Figure 3). Additional to the deepening data on the goat milk composition, a study of chemical composition through X-Ray Fluorescence Spectroscopy by dispersive energy on a Shimadzu equipment, model EDX – 800HS was realized (Table 5).

Physicochemical analysis

The average results of fat, protein, lactose and total solids dry extract (Chart 5.6) are inside the general patterns for the other goat milks, with 3.7% of fat, 3.61% of protein, 4.46% of lactose and 12.17% of total solids dry extract (Table 6). They do not differ much from the values of Pereira et al. (2009). Oliveira (1986) described the centesimal composition of milk of many species, in which the caprine milk contains 4.0% of protein, 3.0% of fat, 4.8% of lactose, 0.8% of ashes and 87.4% of water. Devendra (1972) and Jenness (1980) studied the content of Anglo Nubian milk in tropical and subtropical regions and found the following average content: ESST 12.2%, 4.1% of fat, 4.4% of crude protein and 0.79% of ashes.

Pereira et al. (2009) obtained in the dry season on IFMG, campus Bambui, three repetitions to “in natura” goat milk average levels of fat (3.19%), protein (3.24%), lactose (4.00%), total solids (11.28%) and defatted dry extract (8.09%).

Kouri et al. (2019) obtained mean rates of fat, proteins, lactose and SNF as 34.9 ± 1.8, 38.9 ± 0.6, 48.8 ± 0.8 and 107.1 ± 1.5 g L^{-1} respectively. The concentration of fat dropped by 54.8 % from the 1st (66.5 ± 15.4 g L-1) to the3rd week post-partum (30.0 ± 1.7 g L^{-1}), then stabilized for the rest of early lactation period. Proteins, lactose and SNF rates were the highest in the 1st week post-partum (44.7 ± 4.0, 55.9 ± 4.9 and 122.1 ± 10.0 g L^{-1} respectively). They decreased through the 1st month of lactation before stabilizing in the two next months. The average concentration of minerals remained relatively stable throughout early lactation; ranging between 5.9 and 7.6 g L^{-1} with a mean of 6.7 ± 0.2 g L^{-1}.

The fat results suggest anon-significant increase ($p > 0.05$) by the Tukey test to the treatment 01 with supply of 0.1 mg animal Se.kg^{-1} MS.day^{-1}regarding the control group. However, a significant decrease in the fat content of the group from treatment 02, supplied 0.45 mg animal Se.kg^{-1} MS.day^{-1}($p < 0.05$) were observed, by the Tukey test in relation to the control group (Figure 4).

Gomes et al. (2004) observed during the eight months of lactation that the fat values increased on the first four months, getting to 5.39, from then the percentages decreased. The average value of fat was 4.10% for the

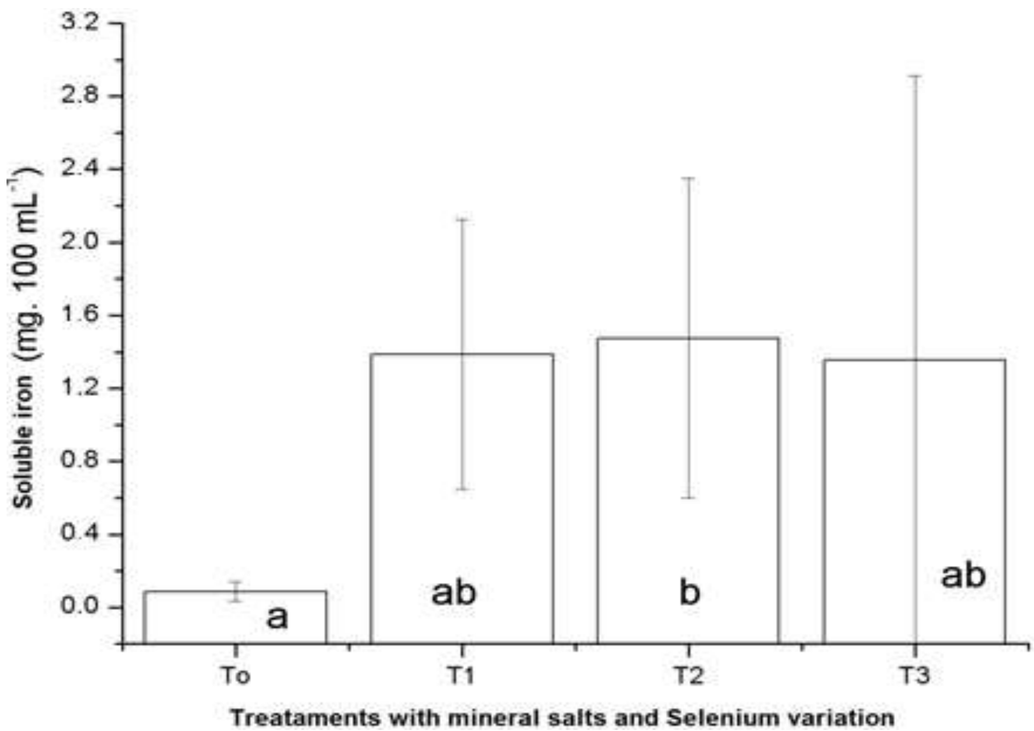

Figure 3. Soluble iron levels on goat milk with adding of mineral salt and three levels of Selenium. The bars represent the averages and the vertical lines the standard errors (n=32). Different letters represent significant difference related to the control group ($p<0.05$) by the Tukey test.

Table 5. Chemical composition of the goat milk from the experiment animals with three different levels of selenium on the diet (n=8), (treatment 00) on the levels **0.0 mg** of animal Se.kg^{-1} DM.day^{-1}obtained through X-Ray Fluorescence Spectroscopy by dispersive energy on a Shimadzu equipment, model EDX – 800HS.

Element	Precentage	μg.cm^{-2}	kα (Alpha layer)
K	36.548	0.588	0.0438
Cl	25.972	0.905	0.0099
Ca	25.593	0.292	0.1193
P	4.227	0.169	0.0095
Cu	3.142	0.196	0.0972
S	1.871	0.100	0.0098
Br	1.817	0.089	0.0233
Fe	0.831	0.084	0.0086

Table 6. Average of fat, protein, lactose and total solids values of goat milk on the period between July and September of 2009, Campus II UNEAL, Santana do Ipanema, Alagoas, 2009

Parameter	Average (%)*
Fat	3.07
Protein	3.61
Lactose	4.46
Dry extract total solids	12.17
* (n=32)	

eight months studied. Studies developed with animals of the Alpina type by Voutsinas et al. (1990) showed average fat values of 3.44%. Bueno et al. (1991), in an experiment realized on 40 Anglo Nubian goats, found values of 4.79% of fat. The percent of lactose results on the goat milk showed significant difference ($p < 0.05$) between the treatments T2 provided 0.45 mg animal Se. kg^{-1} MS. day^{-1} e and the control group treatment T0 with levels 0.0 mg of animal Se.kg^{-1} MS.day^{-1} (Figure 5).This suggests a proportional increase of lactose from the raise of selenium levels on the animal diet.

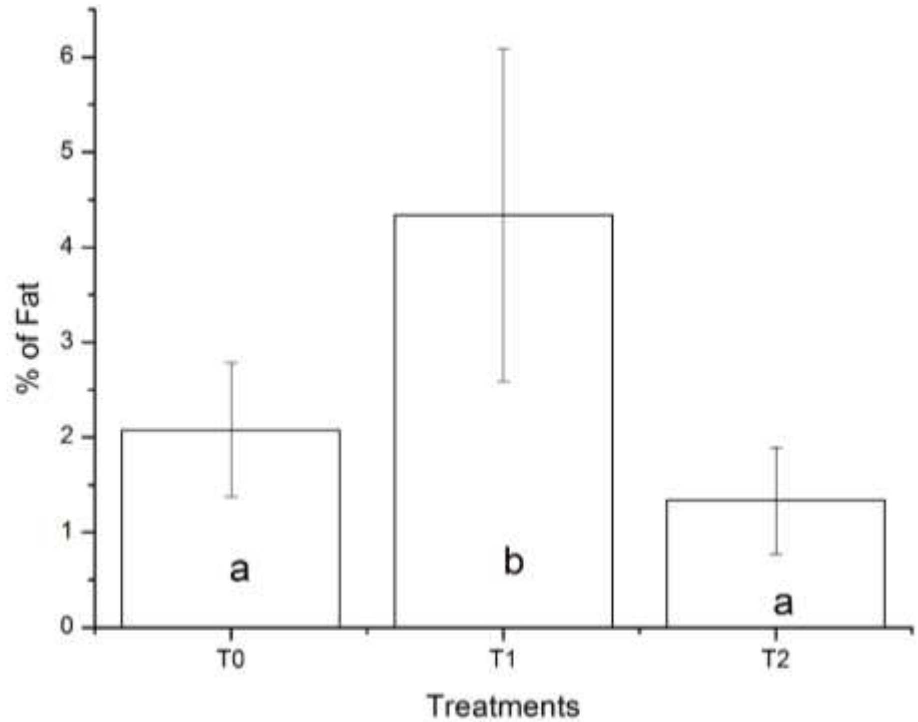

Figure 4. Percentage of fat on goat milk from adding mineral salt and two levels of selenium. The bars represent the averages and the vertical lines the standard errors (n=32). Different letters represent significant difference related to the control group ($p<0.05$) by the Tukey test.

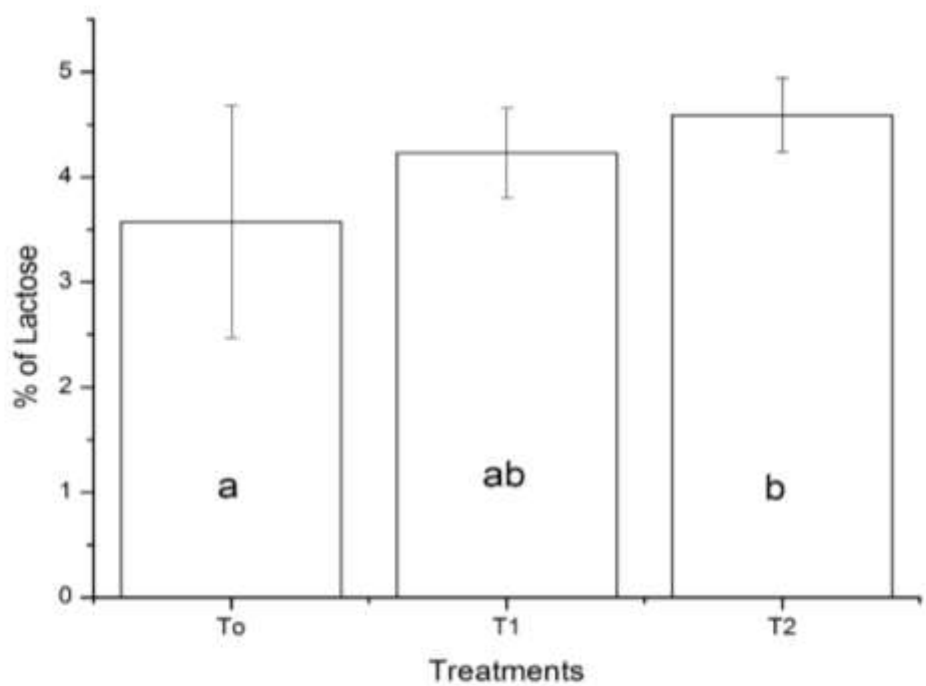

Figure 5. Lactose percentage on the goat milk with adding of mineral salt and two levels of selenium. The bars represent the averages and the vertical lines the standard errors (n=32). Different letters represent significant difference related to the control group ($p<0.05$) by the Tukey test.

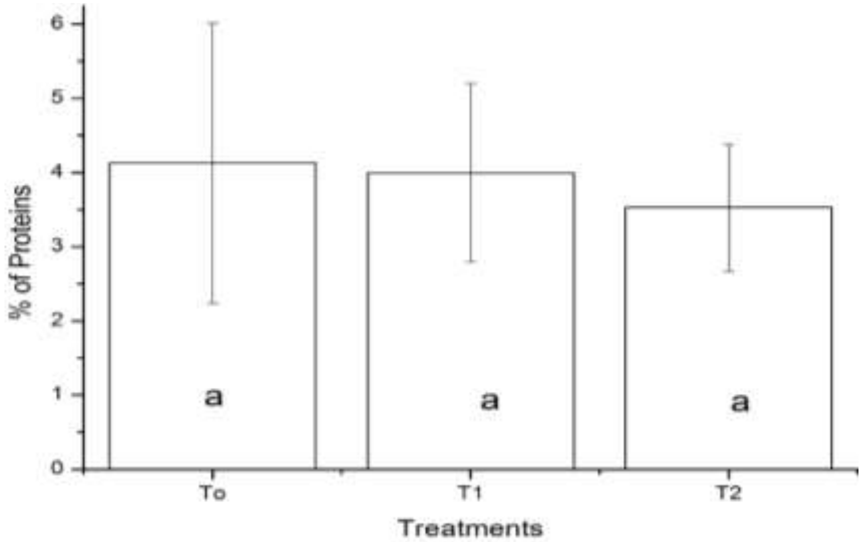

Figure 6. Protein percentage on the goat milk with adding of mineral salt and two levels of selenium. The bars represent the averages and the vertical lines the standard errors (n=32). Different letters represent significant difference related to the control group (p<0.05) by the Tukey test.

Brasil et al. (2000) found that goats under caloric stress, in the morning, have an average of 4.72% of lactose and, in the afternoon, an average of 4.57%. The study shows that the protein results were similar to the control group, not having present significance ($p > 0.05$). Besides that, the results are in good protein levels beside the values presented by others authors. The result obtained in the afternoon equals to that obtained on treatment 2 of thisstudy. Bueno et al (1991), in a study realized on 40 Anglo Nubian goats, found values of 5.32% to the lactose. The protein values resulted from the two treatments with selenium obtained non-significant results ($p > 0.05$).This showed the non-influence of the mineral on protein variation of the milk (Figure 6).

Anglo Nubian goats were studied by D'Alessandro (1991) and showed an average of 4.3% for total protein. Bueno et al (1991), in an experiment realized with 40 Anglo Nubian goats, found values of 3.28% to the protein. From studies with Saanen goats, Prata et al. (1998) concluded that, by the determined global values, 90.83% of the total nitrogen corresponds to the true protein (TP) and 9.17% to the non-protein nitrogen fraction (NNP). The same way, of the true protein, 81.82% corresponds to the casein fraction, important in obtaining dairy products; and 18.18% corresponds to the other proteins remaining afterthe caseins precipitation. From the ESST obtained results, that verify non-significant values ($p > 0.05$) by the Tukey test, on treatment 01 (0.1 mg animal Se. kg^{-1} MS. day^{-1}), there was an addition in relation to the control group. This is related to the elementary fact of the simple supplementation of animals with the commercial mineral salt, also advantageous to the raise of total solids, which would allow a bigger exploitation by volume of milk used on the cheese and other dairy products production (Figure 7); however, non-significant ($p > 0.05$).

Prata et al. (1998), in his studies on Saanen goats, concluded that ESST varied from 10.60 to 15.30%, with 75% of the results up to 13.85% and average of 12.445±0.785%. EESD varied from 8.21 to 10.06%, with 75% of the results up to 9.36 and average of 8.895 ± 0.337%. The average somatic cells count (CCS) to the treatments (treatment 00) on the levels of 0.0mg of animal Se.kg^{-1} MS.day^{-1}; (Treatment 01) with supply of 0.1 mg of animal Se.kg^{-1} MS.day^{-1}. According to Andriguetto (1983 recommendations, (Treatment 02) supplied 0.45 mg of animal Se.kg^{-1} MS. day^{-1} and (Treatment 03) with 0.90mg of animal Se.kg^{-1} MS.day^{-1} was around 409.11 SCC.mL^{-1}.1000, (n=32) and also the average of colony forming units (UFC) was 544.56 UFC.ml^{-1} 1000 (n=24) (Table 7).

As regards the selenium adding treatments to the mineral salt, results only for the treatments 01 and 02 was obtained, because the treatments 03 on the samples were lost. That way, there was a decrease on SCC.mL^{-1}.1000 with significant difference ($p<0.05$) between the control group and the treatment 2 with adding of Se to the mineral salt (Figure 8). Studies developed by Pereira et al. (2009) showed that for the goat milk in the dry season of IFMG, campus Bambui, 2009, the somatic cell count.ml^{-1}.000 showed a result of 1.731 and the colony forming units.ml^{-1}.1000 were 359.

According to Paes et al. (2003), the antioxidants have the function of stopping that there is an accumulation of oxygen reactive species on the cell mean, minimizing compromising damages on the defense cells of the mammary gland. Burk et al. (2008) affirmed that the Se

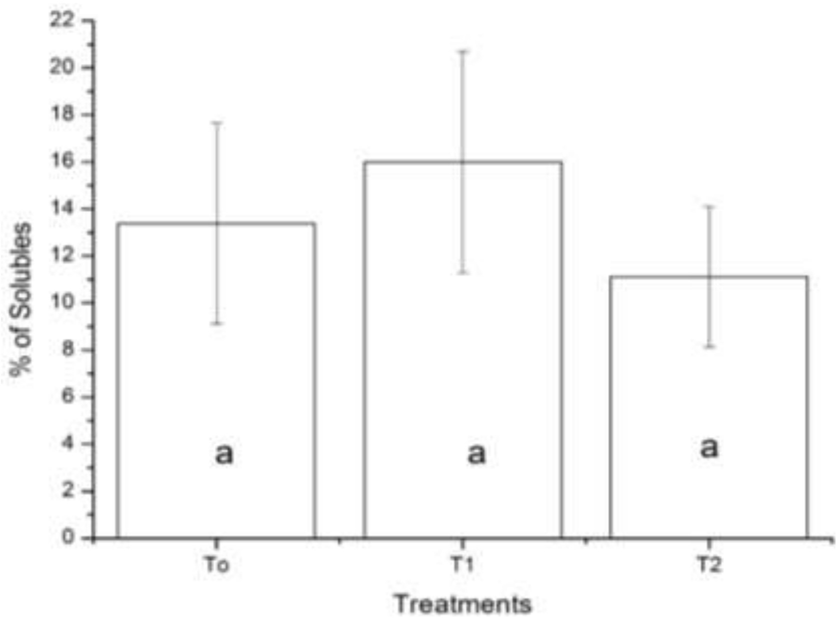

Figure 7. ESST percentage on the goat milk with adding of mineral salt and three levels of selenium. The bars represent the averages and the vertical lines the standard errors (n=32). Different letters represent significant difference related to the control group ($p<0.05$) by the Tukey test.

Table 7. Average values of SCC.ml^{-1}.1000 and UFC/mL^{-1}1000 of goat milk from the period of July to September, Campus II UNEAL, Santana do Ipanema, Alagoas, 2009.

Parameter	Average
CCS.ml^{-1}.1000	409.11*
UFC/ml^{-1}.1000	544.56**
* (n=32) ** (n=24)	

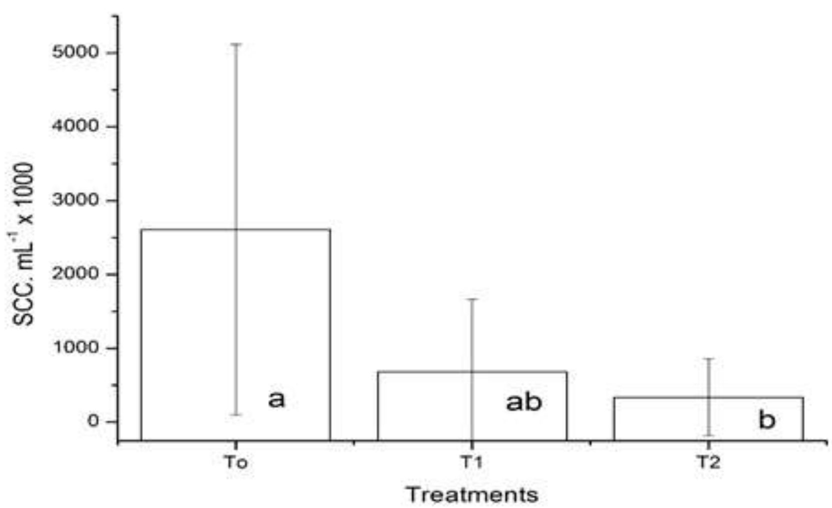

Figure 8. Somatic cell count SCC ml*1.000 on the goat milk with adding of mineral salt and three levels of selenium. The bars represent the averages and the vertical lines the standard errors (n=32). Different letters represent significant difference related to the control group ($p<0.05$) by the Tukey test.

deficiency increases the endogenous oxidative cytosolic stress. The antioxidant action of Se associated to the glutathione peroxidase enzyme works on the metabolism of compounds strange to the organism (Pizzino et al., 2017).

Studies developed in dairy cows by Zanetti et al. (1998) did not show decrease of subclinical mastitis diagnosed through the CMT Test on cows receiving vitamin E, but found a positive effect of Se after implementation with 5mg day^{-1}. The Se and vitamin E supplementation realized in the peripartum on dairy cows by Paschoal et al. (2006) did not affect the milk somatic cell count. This was possibly due to the low levels utilized. The experimental units were distributed randomly in four treatments: 2.5mg of selenium in the form of sodium selenite, 1000 UI of vitamin E in the form of alpha tocopherol acetate, 2.5mg of Se + 1000 UI of vitamin E and the control group. The author suggested new studies to best evaluate the effects of minerals and vitamins supplementation on the prevention and control of mastitis.

Conclusion

The supply of mineral salt proved to be essential in the response of essential minerals to human health, such as Ca and soluble Fe with significant increases, P did not respond to the increase, even decreasing its value with the increase of Selenium. The CCS decreased significantly with the increase of Se in the diet, more extensive studies with different dosages and forms of application are suggested. The Milk did not directly transfer the increased selenium dosages in animal feed. Studies on selenium dosages via venal are suggested to analyze its transfer.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Andriguetto JMM (1983). Nutrição Animal. 2. ed. São Paulo: Nobel, 395p.

Brasil LHA, Wechesler FS, Baccari JF, Gonçalves HC (2000). Efeitos do estressetérmicosobre a produção, composiçãoquímica do leite e respostastermorreguladoras de cabras da raçaAlpina. RevistaBrasileira de Zootecnia 29(6):1632-1641.

Burk RF, Hill KE, Nakayama A, Mostert V, Levander XA, Motley AK, Johnson DA, Johnson JA, Freeman ML, Austin LM (2008). Selenium deficiency activities mouse liver Nrf2-ARE but Vitamin E deficiency does not. Free Radical Biology and Medicine 44(8):1617-1623.

D'alessandro WT (1991). Teor de proteína do leite de cabrasPardaAlpina e Anglo-nubiana. ReuniãoAnual da SBZ, 28.João Pessoa, Anais.

JennessR (1980). Composition and Characteristics of Goat Milk, Review 1968-1979. Journal of Dairy Science 63(10):1605-1630.

DevendraC (1972). The composition of Milk of British Alpine and Anglo-Nubian goats imported into Trinidad. Journal of Dairy Science 39(3):381-385.

El-ramady HR, Domokos-Szabolcsy E, Tarek A. Shalaby A, FáriMG (2015). Selenium in Agriculture: Water, Air, Soil, Plants, Food, Animals and Nanoselenium. In book: Environmental Chemistry for a Sustainable World 5(CO2 sequestration, biofuels and depollution). Chapter: 5. Publisher: Springer Berlin. Editors: E. Lichtfouse DOI: 10.1007/978-3-319-11906-9_5

Ferreira ALA, Matsubara LS (1997).Radicaislivres: conceitos, doençasrelacionadas, sistema de defesa e estresseoxidativo.Revista da Associação Médica Brasileira 43(1):172-173.

Gomes V, Paiva AMM, Libera D, Madureira KM, Araújo WP (2004).Influence of lactation stage on goat (Capra hircus) Milk composition. Brazilian Journal of Veterinary Research and Animal Science 41(1):339-342.

Greenberg AE, Clesceri LS, Eaton AD (2005) Standard methods for the examination of water and wastewater, 21.ed. American Water Works Association.22th edition. 1325 p. ISBN-10: 0875532357 & ISBN-13: 978-0875532356.

Ghany-Hefnawy AE, Tortora-Perez JR (2010).The importance of selenium and the effects of its deficiency in animal health. Small Ruminant Research 89(2–3):185-192.

Hunter RA, Peter DW, Quinn MP, Siebert BD (2005).Intake of selenium and other nutrients in relation to selenium status and productivity of grazing sheep. Australian Journal of Agricultural Research 33(3):637-647.

Kurutas EB (2016). The importance of antioxidants which play the role in cellular response against oxidative/nitrosative stress: current state. Nutrition Journal 15(1):71.

KouriF, Charallah SK, Amina A, Zaina KF (2019). Milk production and its relationship with milk composition, body and udder morphological traits in Bedouin goat reared under arid conditions. ActaScientiarum: Animal Sciences 41:e42552.

Leite L, Oliveira S, Fabiana O, Garcia RCM (2017). Indicators to assess goat welfare on-farm in the semiarid region of Brazilian Northeast. Ciência Rural 47(9):e20161073.

Nascimento EE, Morais GV, Macedo FAF (2008). Suplementação de selênionadieta de caprinossobre a produção e qualidade do embrião. PUBVET 2(23). Available at: http://www.pubvet.com.br/texto.phd?id=251

NRC – National Research Council (2001).Nutrient requirement of the dairy catle. 7. ed. Washington, D.C.

Oliveira JS (1986).Queijo: fundamentostecnológicos. 2. ed. Campinas. UNICAMP 146p.

Powers KA, DhamoonAS (2019). Physiology, Pulmonary, Ventilation and Perfusion.https://www.ncbi.nlm.nih.gov/books/NBK539907/

Paes PRO, Lopes STA, Lopes RS, Kohayagawa A, Takahira RK, Langoni H (2003). Efeitos da administração de vitamina E nainfecçãomamária e nacontagem de célulassomáticas de cabrasprimíparasdesafiadasexperimentalmente com Staphylococcus aureus. ArquivoBrasileiro de MedicinaVeterinária e Zootecnia 55:15-20.

Paschoal JJ, Zanetti MA, Cunha JA (2006). Contagem de célulassomáticas no leite de vacassuplementadas no pré-parto com selênio e vitamina E. Ciência Rural 36:5. DOI: 10.1590/S0103-84782006000500018.

Pereira ED, Duarte NetoJP, PaciulliSOD (2009).Produção e qualidade do leite de cabraproduzido no iFMG- campus Bambuidurante o período das secas. In. II Semana De Ciencia E Tecnoilogia Do Ifmg Campus Bambui: Ii Jornada Cientifica. Bambui-MG. Anais.IFMG.

Pizzino G, IrreraN, CucinottaM, PallioG, ManninoF, ArcoraciV, SquadritoF, AltavillaD, BittoA. 2017. Oxidative Medicine and Cellular Longevity. Available at: https://doi.org/10.1155/2017/8416763. Accessed: 25 mar 2020.

PrataLF., Ribeiro AC, Rezende KT, Carvalho MRB, Ribeiro SDA, Costa, RG(1998). Composição, perfilnitrogenado e caracterís-ticas do leitecaprino (saanen): regiãosudeste, Brasil. Ciencia e Tecnologia de AlimentosCampinas 18:4. DOI: 10.1590/S0101-20611998000400014.

Silva MLS, Vitti GC, Trevizam AR (2007). Concentração de metaispesadosemgrãos de plantascultivadasem solo com diferentesníveis de contaminação. Pesquisa Agropecuária Brasileira, 42(4):527-535.

Souza PT, Salles MGF, Costa ANL, Carneiro HAV, Souza LP, Rondina

D, Araújo AA (2013). Physiological and production response of dairy goats bred in a tropical climate. International Journal of Biometeorology 58:1559–1567. DOI 10.1007/s00484-013-0760-x
Tortuga (2016). Caprinofós. Available at: http://www.avipec.com.br/v2/index.php?option=com_content&view=article&id=398:caprinofos-com-mineraisorganicos&catid=56:tortuga&Itemid= 130.
Voutsinas L, Pappas C, Katsiari M (1990). The composition of Alpine goat's milk during lactation in Greece. Journal of Dairy Research 57(1):41-51.
ZanettiMA,Neunhaus LED, Schalch ED, Martins JH (1998). Efeito da suplementação de Selênio e vitamina E embovinosleiteiros. RevistaBrasileira de Zootecnia 27(2):405-408.

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Inoculation of native arbuscular mycorrhizal fungi based bio-fertilizers for improvement of maize productivity in Central Benin

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The response of maize to a composite inoculation of Arbuscular Mycorrhizal Fungi (AMF) in fields in Central Benin was evaluated. The 2000 SYNEE maize variety underwent 3 AMF groups involved in treatments namely: T1 = "Control (AMF and fertilizer free)"; T2 = "*Glomeraceae*"; T3 = Acau "*Acaulosporacea*"; T4 = Diversi " *Diversisporaceae*"; T5 = "50% NPK + Urea"; T6 = "*Glomeraceae* + 50% NPK + Urea"; T7 = "*Acaulosporaceae* + 50% NPK + Urea"; T8 = "*Diversisporaceae* + 50% NPK + Urea"; T9 = "100% NPK + Urea". The 2 factors were combined in a split-split plot design with a complete randomized block layout including 3 repetitions. The main and subplot factors were respectively the AMF type and use of fertilizers. The growth parameters (height, noose diameter and leaf area), grain yield and endomycorrhizal infection of plants were evaluated. T6 had induced the highest height, fresh above, underground biomass and grain yield of maize passing respectively from 42.38, 25.58, 58.33 and 27.18%, values recorded by the control plants. Moreover, T7 held the highest values of noose diameter and T8 those of mycorrhization intensities. However, T2 and T4 generated the highest values of mycorrhization frequencies and spore number. These results highlighted the advantages of endogenous AMF for sustainable agriculture in Benin.

Key words: Bio-fertilizers, mycorrhizal, composite, maize, sustainable agriculture, Benin.

INTRODUCTION

Arbuscular mycorrhizal fungi (AMF), beneficial microbes to agricultural and natural ecosystems (Pellegrino et al., 2015), improve the host plant's (P) nutrition, growth and resistance to drought and disease (Kumari et al., 2019).

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Thus, AMF present in most soils and colonizing the roots of the majority (80%) of plants (Walder and van der Heijden, 2015) have received particular attention in relation to their application in sustainable agriculture (Chen et al*.,* 2018). Under natural conditions, these obligatory symbionts, unlike ectomycorrhizal symbionts, do not have host specificity and in many cases, a differential effect in each species has been observed with different natures of mycorrhizal inoculum (Diallo et al*.,* 2016; Kouadio et al*.,* 2017). Indeed, Van der Heijden et al*.* (1998) and Abbas (2014) have shown that the benefits of a fungus or AMF complex are not identical within the same plant species (Anguiby et al*.,* 2019). Mycorrhization also increases plants' performance by improving growth (Pavithra et al*.,* 2018), water status and nutrient accumulation (Kapoor et al*.,* 2013). Consequently, increases in yields, particularly for maize and wheat, were highlighted (Treseder, 2013; Pellegrino et al*.,* 2015). During this process, mycorrhizal colonization promotes the establishment of extensive hyphae networks and the secretion of glomaline, which in turn contributes to the absorption of water and nutrients (Pagano, 2014), thus; contributing to a better soil structure (Miller and Jastrow, 2010).

However, under intensive agricultural conditions, AMF have a less significant effect than under natural conditions. The reduction in the number of plant species found in a limited geographical area, such as wheat-corn rotation (Sasvári 2017), regular soil disturbance (Balogoun, et al*.,* 2013), excessive use of mineral fertilizers and fungicide use (Jin et al*.*, 2013) inexorably lead to a decline in the number and activity of AMF (Bakonyi and Csitári, 2018). In areas of constantly exploited land, the number of AMF propagules is seriously reduced (Gottshall et al., 2017). Also, the inadequacy of the fungus introduced under new edaphic conditions sometimes leads to its disappearance (Duponnois et al*.,* 2013; Plenchette et al*.,* 2000). However, Bakonyi and Csitári, (2018) noted that the effectiveness of AMF inoculation depended on soil nutrient availability, type of tillage, fertilizer applied, use of protective agents (especially fungicides) and climate. It therefore appears necessary to study the mycorrhizal crops status and identify the indigenous mycorrhizal complex associated with them (Tchinmegni et al*.,* 2016) in order to integrate this technology into agricultural operations as long as it has been proven that the effects of mycorrhizal symbiosis in a controlled environment (greenhouse) differ from those obtained in a real environment (field) (Hoeksema et al*.,* 2010). For example, Lekberg and Koide (2005) demonstrated that the AMF effects are negligible in real environments than in greenhouses. According to this observation, Bakonyi and Csitári (2018) insisted on the size of the experiment, which must go beyond that of experiment in a controlled environment for fear to be irrelevant.

Thus, this study proposes to evaluate the maize response to composite inoculation of AMF in real conditions. Its objective is to promote the use of mycorrhizal fungi in the technical itinerary of maize production. Moreover, it consists of: assessing the growth parameters of maize after inoculation of a few endogenous AMF and measuring their mycorrhization criteria to determine the best combination.

MATERIALS AND METHODS

The 2000 SYN EE-W maize variety was used during the experiment. These are 75-day early varieties, developed by the International Institute of Tropical Agriculture (IITA) and the National Institute of Agricultural Research of Benin (INRAB) (MAEP, 2017).

Determination of soil chemical parameters

Soil samples were collected at a depth of 0-20 cm (Adjanohoun et al*.,* 2011). Sampling consisted of 500 g composite samples at the level of each plot before the test was set up just after ploughing and before any application of fertilizer amendments. These samples were sent to the Soil Sciences, Water and Environment Laboratory (LSSEE) of the National Institute of Agricultural Research of Benin (INRAB). These analyses consisted of the determination of pH (water) (by glass electrodes in a soil-to-water ratio of 1/2.5), organic carbon (Anne, 1945), assimilable P (Bray and Kurtz, 1945), total N (Kjeldahl, 1883), and exchangeable bases using the Metson (1956)'s method with ammonium acetate at a pH of 7.

Isolation of AMF

The groups of mycorrhizal fungi *Glomeraceae* (*Funeliformis mosseae*, *Funeliformis geosporum*, *Glomus caledonius*, *Glomus ambiosporum*, *Rhizophagus intraradices* and *Septoglomus contrictum*), *Acaulosporaceae* (*Acaulospora capsicula, Acaulospora denticulata*) and *Diversisporaceae* (*Diversispora globifera*) were identified from the maize rhizosphere of the 6 different municipalities in the department of Collines using the Davis- INVAM key. These different groups of fungi were isolated and multiplied over a period of twelve weeks using sorghum as a trap plant because of its high mycorrhizogenic potential and as a substrate clay + sterile soil at respective doses of 3⁄4 and 1⁄4 in a 250 ml pot.

Trapping and preparation of inoculums

Sorghum seeds were previously soaked for 2 min–in a 0.024% sodium hypochlorite (NaClO) solution and rinsed 5 times with distilled water under vortex shaking (Gholami et al*.*, 2009). Then, 10 sorghum seeds (*Sorghum bicolor* L.) were sown in each pot containing different levels of soil substrate dilution. The pots were then placed in a greenhouse at room temperature. The plants were watered daily with (tap) water to maintain soil capacity similar to that in the field for 6 weeks. The root biomass and substrate were therefore crushed to obtain the inoculums.

Seed coating

The inoculums obtained were used to coat the seeds in a 10:1 ratio (10 kg of seed per 1kg of bio-product) for each group of fungi. Each mixture was mixed with a quantity of water equivalent to 600 $ml.kg^{-1}$ of fertilizers. The coated seeds were dried in ambient air for 12 h in accordance with the recommendations published by Fernández et al. (2000).

Experimental design

The trial was installed at the Miniffi Research and Development (RD) station in the municipality of Dassa-Zoumé in Benin. The choice was based on the decline of soil fertility. Flat ploughing was carried out the day before sowing on a not flooded plot with a slope of at most 2%. Each plot had an area of 12.8 m² and consisted of 4 lines of 4 m long with a spacing of 0.80 m. Seeding was done at a spacing of 0.80 m x 0.40 m with a density of 31,250 plants/ha (Yallou et al., 2010). The distances between the plots and the repetitions between them were 1.8 m and 2 m, respectively. The useful plot had an area of 6.4 m^2, where data were collected on the 2 central lines. The different treatments are: T1 = "Control (no mycorrhizal fungus and no P addition)"; T2 = "*Glomeraceae*"; T3 = Acau "Inoculation with *Acaulosporaceae*"; T4 = Various "*Diversisporaceae*"; T5 = "50%NPK+Urea"; T6 = "*Glomeraceae* + 50%NPK+Urea"; T7 = "*Acaulosporaceae* +50%NPK+Urea"; T8 = "*Diversisporaceae+50%NPK+Urea*"; T9 = "100%NPK+Urea". Two factors were combined in a split-split plot system (split-split plot with a complete randomized block layout) with 3 repetitions; the large plot factor was the mycorrhizal fungus type and the subplot factor was the use of chemical fertilizer. Mineral fertilizer rate recommended by INRAB (1995) was 200 kg/ha NPK.

Sowing and maintenance of plots

Two coated or uncoated maize (*Zea mays L.*) seeds were sown 5 cm deep. The demarcation to one plant per hole was done at 7th DAS (Day after sowing). The various parameters were collected every 15 days from the 15th to the 60th DAS. Only the leaf area was measured at the 60th DAS.

Evaluation of growth parameters

The effects of the different treatments on maize growth were assessed by measuring the plant height, noose diameter, number of leaves and leaf area. The height of maize plant is the distance between the noose and the last ligule. It was measured with a measuring tape, while the noose diameter was measured with a caliper every 15 days, the 15th, 30th, 45th, 60th DAS. The area of the leaves with ligule was estimated by multiplying the product of leaves length (from the top of the sheath to the tip of the blade) and width (measured in the middle of the blade) by the coefficient 0.75 (Ruget et al., 1996). This measure concerned the last 2 leaves with ligule of each plant and was evaluated at the 60th DAS.

Evaluation of performance parameters

Biomass produced

The determination of above and underground biomass yield was determined as:

$$\boldsymbol{R}' = \frac{\boldsymbol{P}' \times \mathbf{10.000}}{\boldsymbol{S}' \times \mathbf{1.000}}$$

Where: R '= average yield of dry biomass of maize plants t. ha-1,P = weight of dry biomass of maize plants in kg, 10000 is the conversion of hectare in m²; 1000 converting tonne (t) into kg, S = crop area cultivated in m².

Maize grain yield

The harvested maize cobs were weighed per treatment using a precision scale. The moisture content of the grains was determined using a moisture meter (LDS-1F). The average of maize grain yield was determined by Valdés et al. (2013):

$$\boldsymbol{R} = \frac{\boldsymbol{P} \times \mathbf{10.000}}{\boldsymbol{S} \times \mathbf{1.000}} \times \frac{\mathbf{14}}{\%\,\boldsymbol{H}}$$

R = Average yield in maize grains (t. ha^{-1})' P = weight of maize grains in kilograms (kg) ; 10000 is the conversion from ha to m^2; 1000 is the conversion from tonne (t) to kg; S = harvest area in m²; % H = percentage of grain moisture in %.

Evaluation of endomycorrhizal infection of plant roots

At 40 DAS, maize root samples were collected. After staining the roots with trypan blue according to the method described by Phillips and Haymain (1970), root fragments from maize plants were observed in binoculars (XSP-BM-2CEA .2013). Estimation of mycorrhizal root infection was performed using the intersection method (Giovannetti and Mosse, 1980; Trouvelot et al., 1986). Two parameters of arbuscular mycorrhizal infections were used to calculate the mycorrhization rate: The frequency of mycorrhization (F) which reflects the degree of infection of the root system:

$$\mathbf{F\ (\%)} = \frac{(\mathbf{N} - \mathbf{n_o})}{\boldsymbol{N}}$$

Where N corresponds to the number of fragments observed and no is the number of fragments without any trace of mycorrhization.

The mycorrhization intensity: m (absolute mycorrhization intensity) which expresses the portion of the colonized cortex relative to the entire root system:

$$\mathbf{m}(\%) = \frac{\mathbf{95n_5} + \mathbf{70n_4} + \mathbf{30n_3} + \mathbf{5n_2} + \mathbf{n_1}}{\mathbf{N} - \mathbf{n_o}}$$

In this formula, $\mathbf{n_5}$, $\mathbf{n_4}$, $\mathbf{n_3}$, $\mathbf{n_2}$ and $\mathbf{n_1}$ are the numbers of fragments respectively noted in the five infection classes indicating the importance of mycorrhization namely: 5 = more than 95%, 4= from 50 to 95%, 3= 30 to 50%, 2 = 1 to 30%, 1= 1% of the cortex.

Data analysis

All analyses were performed with software R (3.5.3) (R Core Team 2018). A repeated measurement variance analysis was performed on the height, diameter and values related to leaves with the nlme (Linear and Nonlinear Mixed Effects) package (Pinheiro et al., 2019). For Fresh Above Biomass (FAB), Leaf Area (LA) and fresh underground biomass (FUB), we performed multiple variance analysis.

RESULTS

Soil chemical characteristics

The soil chemical properties of the site before the installation of the tests (Table 1) generally showed that the soil of Miniffi, the centre of Benin was slightly basic

Table 1. Chemical characteristics of the soil at the experimental site.

Site	Village	Depth (cm)	pH (H_2O)	C-org (g/kg)	N-total (g/kg)	P-Bray1 (mg/kg)	Exchangeable bases (cmol/kg)		
							Ca^{2+}	Mg^{2+}	K^{+}
Dassa	Miniffi	0 – 20	7.80	8.0	0.60	47.50	3.32	2.31	2.21

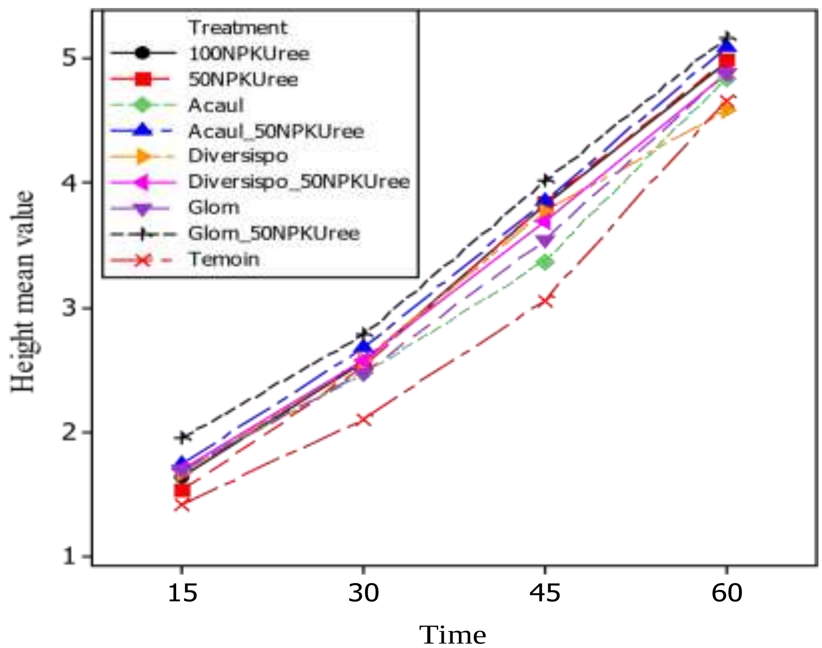

Figure 1. Variation in maize plant height with treatment and time. **Glom:** Glomeraceae, Acaul: Acaulosporaceae, Diversispo: Diversisporaceae, Glom 50 NPKUrée: Glomeraceae + 50 NPK + Urée, Acaul 50 NPKUrea: Acaulosporaceae + 50 NPK + Urée, Diversispo 50 NPKUrea : Diversisporaceae + 50 NPK + Urea).

(pH = 7.8) at the horizon, 0-20 cm. The soil showed intermediate fertility and was richer in Ca^{2+} than the potassium (K^{+}) exchangeable cations. The carbon/nitrogen (C/N) ratio was relatively low at the topsoil level. The P concentration in the topsoil (47.5 mg/kg soil) was low.

Influence of AMF on the height and noose diameter of maize plants

Height

Figure 1 shows the evolution of the height depending on strains and time. A significant effect ($p<0.001$) of both strains and treatments was observed. *Glomeraceae-50NPK+Urea* followed by *Acaulosporaceae-50NPkUrea* were the treatments that induced the highest plant values over the entire period of the experiment (Figure 1) with respective increases of 42.38, 36.59% compared to the control.

Noose diameter

The results of the plant analysis revealed that the plants grew in diameter gradually according to the strains and blocks over time with a significant effect ($p<0.001$). The largest diameters were consistently generated by *Acaulosporaceae + 50% NPK + Urea* followed by *Glomeraceae + 50% NPK + Urea* (Figure 2).

Influence of AMF on Biomass produced, leaf area of plants and maize grain yield

Analysis of the results in Table 1 revealed that *Glomeraceae + 50% NPK Urea* induced an important fresh above biomass (FAB) followed by *Acaulosporaceae + 50% NPK Urea* with respective increases of 25.58 and 22.32% over the control. The results of the multiple variance analyses revealed significant differences between treatments for fresh underground biomass (FUB) and dry underground biomass (DUB).

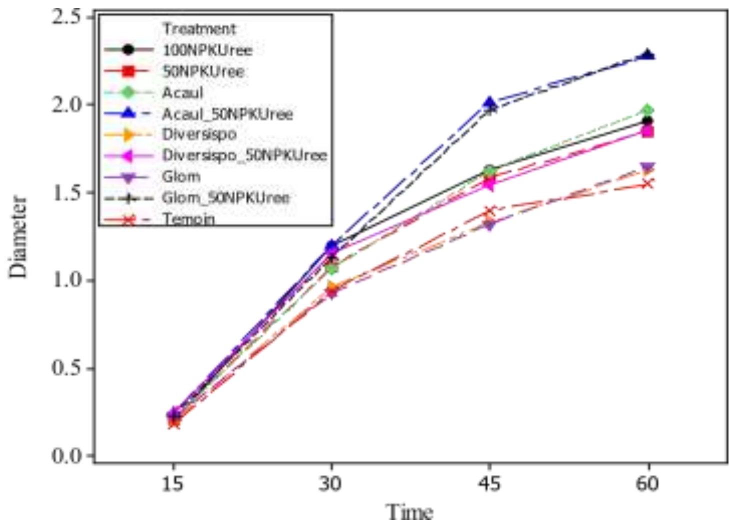

Figure 2. Variation in noose diameter of maize plants depending on treatments and time.

Table 2. Mean values of fresh above biomass, dry above biomass, fresh underground biomass, dry underground biomass, leaf area and yield.

Treatment	FAB	DAB	FUB	DUB	LA	Yield
100 NPK Urea	5.16	4.26	0.45^{ab}	0.36^{ab}	399.22	1.86
50 NPK U rea	4.81	3.48	0.36^{ab}	0.25^{ab}	366.31	2.01
Acaul	5.23	4.05	0.43^{ab}	0.33^{ab}	355.67	2.32
Acaul_50 NPK Urea	5.81	4.21	0.55^{ab}	0.47^{a}	461.33	2.19
Diversispo	4.63	3.31	0.36^{ab}	0.25^{ab}	391.02	1.75
Diversispo_50 NPK Urea	5.27	4.27	0.56^{ab}	0.50^{a}	436.09	1.84
Glom	5.41	4.66	0.37^{ab}	0.27^{ab}	462.59	1.97
Glom_50 NPK Urea	6.07	4.70	0.67^{a}	0.52^{a}	461.40	2.39
Control	4.52	3.18	0.28^{b}	0.18^{b}	314.51	1.73
Probability	0.041	0.054	0.014	0.005	0.142	0.095

Glom: Glomeraceae, Acaul: Acaulosporaceae, Diversispo : Diversisporaceae, Glom 50 NPKUrea : Glomeraceae + 50 NPK + Urea, Acaul 50 NPKUrea: Acaulosporaceae + 50 NPK + Urea, Diversispo 50 NPKUrea : Diversisporaceae + 50 NPK + Urea). FAB: fresh above biomass; DAB: dry above biomass; FUB: fresh underground biomass; DUB: dry underground biomass.

Glomeraceae + 50% NPK Urea generated high value of FUB with an increase of 58.33% compared to control.

Regarding DUB, *Glomeraceae + 50% NPK Urea*, *Diversporaceae* + *50%* NPK+ Urea and *Acaulosporaceae + 50 NPK Urea* have induced a high and similar values. No significant differences were found between yield average and leaf area (LA) mean values. The best grain yield was obtained with *Glomeraceae + 50% NPK Urea* while the largest leaf area was obtained with *Glomeraceae* only*,* exceeding the control by 27.18 and 32.01%, respectively (Table 2).

Mycorrhization parameters

The analysis of the mycorrhization parameters presented in the radar diagram showed that *Diversporaceae* + *50%* NPK+ Urea followed by *Glomeraceae* + 50% NPK + Urea induced the highest mycorrhization intensities unlike *Acaulosporaceae* which generated the lowest intensity (Figure 3a). With regard to mycorrhization frequency (Figure 3c) and spore number (Figure 3b), *Glomeraceae* and *Diversisporaceae* induced the highest values.

DISCUSSION

AMF are soil-borne fungi that can significantly improve plant nutrient uptake and resistance to several a biotic stress (Sun et al*.,* 2018), enabling thereby the establishment of plants in land (Rouphael et al*.,* 2015). However, use of appropriate AMF, either individually or in combination, still remains a big challenge as the benefits

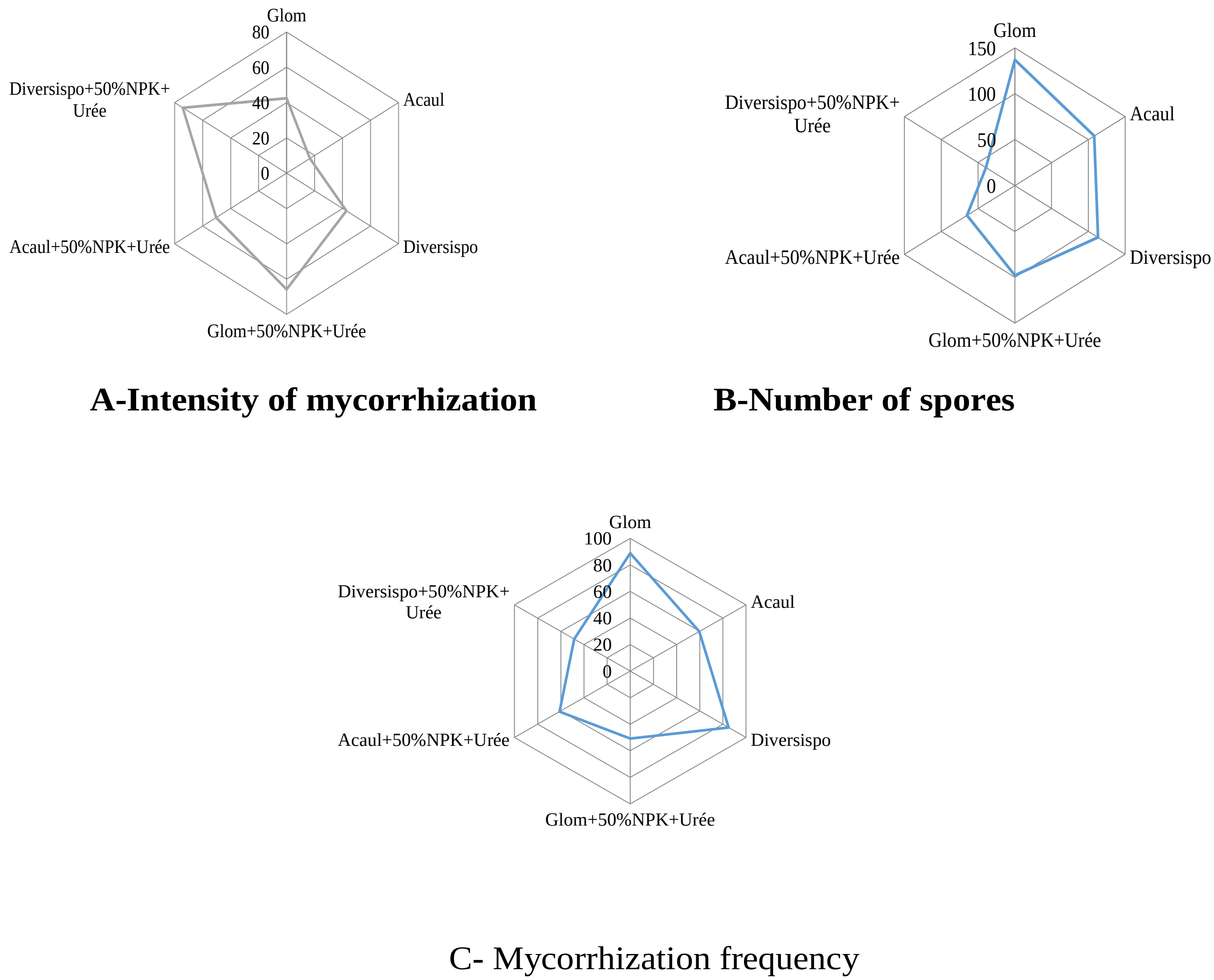

Figure 3. Variation in mycorrhization intensity (a), spore number (b) and mycorrhization frequency (c) depending on treatment (*Glom: Glomeraceae, Acaul: Acaulosporaceae, Diversispo : Diversisporaceae, Glom 50 NPK Urea: Glomeraceae + 50 NPK + Urée, Acaul 50 NPK Urea: Acaulosporaceae + 50 NPK + Urea, Diversispo 50 NPK Urea : Diversisporaceae + 50 NPK + Urea).*

from the mutualism greatly vary depending on the AMF strains (Chen et al., 2017). This study was conducted to evaluate the effect of a composite AMF inoculation on maize in field conditions in Central Benin. The results of the soil samples taken from the site revealed that the soil prior to installation was slightly basic (pH = 7.8) at the horizon of 0-20 cm. The soils have intermediate fertility and are richer in Ca^{2+} than in K^{+} ions. The carbon nitrogen-C/N ratio was relatively low at the topsoil level. Those data are similar to those observed by Igue et al*.* (2015) and the weakness of soil characteristics suggested a good expression of AMF. Hence, the mycorrhization of maize with endogenous strains has significantly enhanced growth parameters such as height, noose diameter, while there is similar effect on leaf area. This improvement may be attributed to both a good mineral uptake of plant (Malonda et al*.,* 2019; Gnamkoulamba et al., 2018) and AMF adaptability to environment traits (Benjelloun, 2014). Benjelloun (2014) proved better that plants of maize inoculated grew better than those not inoculated.

Growth in height, noose diameter at the experimental site was significantly different ($p<0.01$). *Glomeraceae-50NPK+Urea* had induced the greatest heights while the greatest diameters were obtained with *Acaulosporaceae-50NPK-Urea* with respective increases of 42.38, 34.89% compared to the control. Leaf areas of maize plants were not significantly different and the largest leaf area was obtained with only *Glomeraceae,* exceeding the control by 32.01% (Table 2). Balliu et al*.* (2015) supported these results by observing an improvement in leaf area, nitrogen, potassium, calcium and phosphorus levels, indicating an increase in growth of tomato plants. These rates of improvement in the height, noose diameter and leaf area of maize plants are believed to be due to the positive effect of AMF, which might facilitate the absorption of mineral elements by plants (Malonda et al., 2019) and decrease the uptake of Na and Cl (Evelin et al*.,* 2012), leading to growth stimulation of mycorrhizal plants (Gnamkoulamba et al., 2018).

Glomeraceae+50%NPKUrea induced a large FAB and FUB with increases of 25.58 and 58.33% respectively compared to the control. The same trends were observed by Gnamkoulamba et al. (2018a, 2018b) who recorded a significant total dry rice biomass and indicated an increasing trend with the inoculation dose. These results would be due to the significant improvement in the water balance, the absorption of certain trace minerals and macronutrients from the plants by AMF; an increase in the production of photosynthates improves the accumulation of biomass (Nyembo et al., 2012; Mitra et al., 2019) interrelated to photosynthetic performance of a plant (Chen et al*.,* 2017). Zhang et al*.* (2018) demonstrated the AMF involvement in amelioration of the biomass allocated to stems, panicles and grains with an increased redistribution of phosphorus and nitrogen to grains. The effects of AMF used in this study were clearly demonstrated through the improvement of all growth and yield parameters. The best grain yield was obtained with *Glomeraceae+ 50%NPKUrea,* exceeding the Control by 27.18% (Table 2). This should be explained by the production of glomalin-related soil protein containing 30–40% of carbon and its related compounds that safeguard soil from desiccation by enhancing the soil water holding capacity (Sharma et al*.,* 2017). This rate of grain yield improvement was below the 51% obtained by Aboubacar et al. (2013) in cowpea after inoculation of mycorrhizal fungi and slightly above the 20% got by Ceballos et al. (2013) in cassava. These results support the high mycorrhization frequency and intensity and the large numbers of spores obtained. Therefore, Diagne et al*.* (2003) mentioned that host plants got many benefits when intensity of mycorrhisation is above12%. Beltrano et al*.* (2013) and Sarah and Ibrar (2016) suggested that spore density and host colonization are largely defined by the compatibility of AMF with host plants, environment and interactions between AMF and exudates produced by the host plant. In addition, the importance of AMF is also highlighted by the increase in mycorrhizal infection even under hostile conditions. Our study demonstrated that the combinations, *Diversporaceae+50%* NPK+ Urea followed by *Glomeraceae+50%NPK+Urea* had the highest mycorrhization intensities, showing a significant development of fungal mechanisms promoting nutrient exchange between the 2 symbiotic partners (Benjelloun et al*.,* 2014).

Conclusion

This study compared the behaviour of different groups of AMF on maize growth and grain yield variables in field environment. *Glomeraceae50NPK+Urea* induced the highest heights, yield of fresh above-ground and underground fresh biomass with the highest grain yields of maize exceeding the values measured in the control plants by respectively 42.38, 25.58, 58.33% and 27.18%. Regarding the noose diameter, the largest values were measured with *Acaulosporaceae+50%NPK*. These results showed that the mycorrhization of maize with the different strains of used AMF could improve the productivity of this crop by halving the use of mineral fertilizers. Thus, it is therefore imperative to popularize this technology by developing an organic fertilizer based on AMF for sustainable agriculture in Benin.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Abbas Y (2014). Microorganisms in the rhizosphere of Tetraclinaria: a tool to optimize the assisted regeneration of *Tetraclinis articulates* Vahl. M.Sc., Ph.D. Thesis, Mohammed V University, Faculty of Science Rabat, 157p.

Aboubacar K, Zakari MO, Harouna IA, Seydou I, Alzouma M, Zoubeirou (2013). Effect of co-inoculation of rhizobium and mycorrhizae on the agronomic performance of cowpeas (*Vigna unguiculata* (L.) Walp.) in Niger. Journal of Applied Biosciences 72:5846-5854.

Anguiby BLA, Ouattara G, Bomisso EL, N'goran B, Ouattara B, Coulibaly SA, Aké S (2019). Evaluation du statut mycorhizien d'arbres de Ceiba pentandra (L), Gaertn et Tieghemella heckelii (A.Chev), Pierre, du jardin Botanique de Bingerville en Côte d'Ivoire Journal of Applied Biosciences 138:14092-14105.

Anne P (1945). Sur le dosage rapide du carbone organique dans les sold Ann: Agroni Avril, Mai, Juin, 1945, 5he année 2:161-172.

Bakonyi I, Csitári G (2018). Response of winter wheat to arbuscular mycorrhizal fungal inoculation under farm conditions. Journal of Agriculture and Environmental Sciences 5:1.

Balliu A, Sallaku G, Rewald B (2015). AMF Inoculation enhances growth and improves the nutrient uptake rates of transplanted, salt-stressed tomato seedlings. Sustainability 7:15967-15981.

Balogoun I, Saïdou A, Ahoton LE, Adjanohoun A, Amadji GL, Ezui G, Youl S, Mando A, Igué AM, Sinsin BA (2013). Détermination des formules d'engrais minéraux et des périodes de semis pour une meilleure production du maïs (*Zea mays* L.) au Sud et au Centre Bénin. Bulletin de la Recherche Agronomique du Bénin, ISSN sur papier : 1025-2355, Numéro spécial Fertilité du maïs, pp.: 1-25

Beltrano J, Ruscittil M, Arango MC, Ronco M (2013). Effects of Arbuscular Mycorrhiza Inoculation on Plant Growth, Biological and Physiological Parameters and Mineral Nutrition in Pepper Grown under Different Salinity and P Levels. Journal of Soil Science and Plant Nutrition 13(1):123-141.

Benjelloun S, El Harchli EH, Amrani JK, El Ghachtouli N, Fikri Benbrahim K, El Yamani J (2014). Etude De L'importance De la Mycorhization dans la Synthèse des composés phénoliques chez le Maïs (*Zea mays* L.) en condition de stress hydrique. Research Inventy: International Journal of Engineering and Science 4(12):43-49.

Bray RH, Kurtz LT(1945). Determination of total organic and available forms of phosphorus in soils. Soil Science 59:39-45.

Ceballos I, Ruiz M, Fernández C, Peña R, Rodriguez A, Sanders IR (2013). The in vitro mass-produced model mycorrhizal fungus, Rhizophagus irregularis, significantly increases yields of the globally important food security crop cassava. PLOS ONE 8(8):e70633.

Chen S, Zhao H, Zou C, Li Y, Chen Y, Wang Z, Jiang Y, Liu A, Zhao P, Wang M, Ahammed GJ (2017). Combined Inoculation with Multiple Arbuscular Mycorrhizal Fungi Improves Growth, Nutrient Uptake and Photosynthesis in Cucumber Seedlings. Frontiers in Microbiology 8:2516.

Chen MM, Arato L, Borghi E, Nouri, Reinhardt D (2018). Beneficial Services of Arbuscular Mycorrhizal Fungi – From Ecology to Application. Frontiers in Plant Science 9:1270.

Diallo B, Samba SAN, Sane D (2016). Effects of MA fungi on the growth and development of castor seedlings grown under saline stress under semiconductor conditions. Revue des Energies Renouvelables19(1):59-68.

Duponnois R, Ramanankierana H, Hafidi M, Baohanta R, Baudoin E, Thioulouse J, Lebrun M (2013). Endemic plant resources to sustainably optimize forest cover rehabilitation operations in Mediterranean and tropical environments: example of plants that facilitate the spread of mycorrhizal fungi. Biology Reports 336 (5-6):265-272.

Evelin H, Giri B, Kapoor R (2012). Contribution of *Glomus intraradices* inoculation to nutrient acquisition and mitigation of ionic imbalance in NaCl-stressed *Trigonella foenum-graecum*. *Mycorrhiza* 22:203-217.

Fernández F, Gómez R, Vanegas LF, de la Noval BM, Martínez MA (2000). Mycorrhizogenic inoculant product. National Office of Industrial Property. Cuba, Patent No. 22641.

Gholami A, Shahsavani S, Nezarat S (2009). The Effect of Plant Growth Promoting Rhizobacteria (PGPR) on Germination, Seedling Growth and Yield of Maize. World Academy of Science, Engineering and Technology 49:19-24.

Giovannetti M, Mosse B (1980). *An evaluation of techniques for measuring vesicular– arbuscular infection in roots*. New Phytologist 84:489-500.

Gnamkoulamba A, Tounou AK, Tchao M, Tchabi A, Adjevi AKM, Batawila K (2018). Field assessment of the growth potential and production of rice (*Oryza sativa* L.) variety IR841 inoculated in the nursery by four strains of arbuscular mycorrhizal fungi. European Scientific Journal 14:12.

Gottshall CB, Cooper M, Emery SM (2017). Activity, diversity and function of arbuscular mycorrhizae vary with changes in agricultural management intensity. Agriculture, Ecosystems and Environment 241:142-149.

Hoeksema JD (2010). A meta-analysis of context-dependency in plant response to inoculation with mycorrhizal fungi. Ecology Letters 13:394-407.

Igue MA, Oga AC, Balogoun I, Saidou A, Ezui GYS, Kpagbin G, Mando A, Sogbedji JM (2016). Détermination des formules d'engrais minéraux et organiques sur deux types de sols pour une meilleure productivité de maïs (*Zea mays* l.) dans la commune de Banikoara (Nord-Est Du Bénin). European Scientific Journal 12:30.

Jin H, Germida JJ, Walley FL (2013). Suppressive effects of seed-applied fungicides on arbuscular mycorrhizal fungi (AMF) differ with fungicide mode of action and AMF species. Applied Soil Ecology 72:22-30.

Kapoor R, Evelin H, Mathur P, Giri B (2013). Arbuscular mycorrhiza: Approaches for abiotic stress tolerance incropplantsforsustainableagriculture. In Plan tAcclimation to Environmental Stress; Tuteja, N., Gill, S.S., Eds.; Springer: New York, NY, USA,; pp. 359-401.

Kjeldahl J (1883). Neue Methode zur Bestimmung des Stickstoffs in organischen Körpern. Zeitschrift für Analytische Chemie 22:366-382.

Kouadio AN, Nandjui J, Zézé A (2017). An Indigenous and an Exotic AMF Strains Improve Dioscorea Alata growth and induce tolerance to Meloidogyne spp., Agricultural Science Research Journal 7(2):63-69.

Kumari S, Merina P, Srimeena N (2019). Arbuscular Mycorrhizal Fungi (AMF) Induced Defense Factors against the Damping-off Disease Pathogen, *Pythium aphanidermatum* in Chilli (Capsicum annum). International Journal of Current Microbiology and Applied Sciences 8(6):2243-2248.

Lekberg Y, Koide RT (2005): Is plant performance limited by abundance of arbuscular mycorrhizal fungi? A meta-analysis of studies published between 1988 and 2003. New Phytologist 168:189-204.

MAEP (Ministère de l'Agriculture de l'Elevage et de la Pêche) (2017). Recueil des technologies agricoles prometteuses développées par le Système National de Recherche Agricole (SNRA) de 1996 à 2015 Document Technique et d'Informations. ISBN : 978-99919-2-985-9 Dépôt légal n° 9433 du 12 juin 2017 Bibliothèque Nationale du Bénin, 2èmetrimestre. 288p

Malonda AN, Nzola-meso TM, Manga AM, Yandju MC (2019). Effet des champignons mycorhiziens Arbusculaires sur le phosphore des sols tropicaux et implication dans la biosynthèse du caroténoïde du manioc. Journal of Applied Biosciences 135:137.

Miller RM, Jastrow JD (2010). Mycorrhizal fungi influence soil structure. In: Kapulnik, Y., Douds, D. (Eds.) Arbuscular mycorrhizas: Physiology and function. Chapter 1. Kluwer Academic Publishers.

Mitra D, Navendra U, Panneerselvam U, Ansuman S, Ganeshamurthy AN, Divya J (2019). Role of mycorrhiza and its associated bacteria on plant growth promotion and nutrient management in sustainable agriculture. International Journal of Life Sciences and Applied Science 1:1-10.

Pagano MC (2014). Drought stress and mycorrhizal plant. In Use of Microbes for the Alleviation of Soil Stresses; Miransari, M., Ed.; Springer: New York, NY, USA 1:97-110.

Pavithra D, Yapa N (2018). Arbuscular mycorrhizal fungi inoculation enhances drought stress tolerance of plants. Groundwater for Sustainable Development 7:490-494.

Pellegrino E, Opik M, Bonari E, Ercoli L (2015). Responses of wheat to arbuscular mycorrhizal fungi: A meta-analysis of field studies from 1975 to 2013. Soil Biology and Biochemistry 84:210-217

Phillip JM, Hayman DS (1970). Improved procedures for cleaning roots and staining parasitic and vesicular arbuscularmycorrhizal fungi for rapid assessment of infection. Transactions of the British Mycological Society 5:158-161.

Pinheiro J, Bates D, DebRoy S, Sarkar D, R Core Team (2019). nlme: Linear and Nonlinear Mixed Effects Models. R Package Version 31-141 https://CRAN.R-project.org/package=nlme.

Plenchette C, Bois JF, Duponnois R, Cadet P (2000). Mycorrhization (Glomus aggregatum) of millet (*Pennisetum glaucum*). Soil Studies and Management 7(4):379-383.

Rouphael Y, Franken P, Schneider C, Schwarz D, Giovannetti M, Agnolucci M (2015). Arbuscular mycorrhizal fungi act as biostimulants in horticultural crops. Scientia Horticulturae 196:91-108.

Ruget F, Bonhomme R, Chartier M (1996). Estimation Simple de la Surface Foliaire de Plantes de Maïs en Croissance. Agronomien 16(9):553-562.

Sarah S, Ibrar M (2016). Effects of arbuscular mycorrhizal fungi on spores density and root colonization of four hybrids of sunflower (*Helianthus annuus* L.) at different rock phosphate levels. Sarhad Journal of Agriculture 32(4):258-266.

Sasvári Z (2017). Study of arbuscular mycorrhizal fungal diversity in long-term field experiments. PhD dissertation, Szent István University, Gödöllő.

Sharma S, Prasad R, Varma A, Sharma AK (2017). Glycoprotein associated with *Funneliformis coronatum*, *Gigaspora margarita* and *Acaulospora scrobiculata* suppress the plant pathogens *in vitro*. Asian Journal of Plant Pathology 11(4):192-202.

Sun Z, Song J, Xin X, Xie X, Zhao B (2018). Arbuscular mycorrhizal fungal proteins 14-3-3- are involved in arbuscule formation and responses to a biotic stress during AM symbiosis. Frontiers in Microbiology 5:9-19.

Tchinmegni FI, Tsobeng AC, Ngonkeu MEL, Tchoundjeu Z (2016). Evaluation du statut mycorhizien chez Allanblackia floribunda en vue de sa domestication en zones forestières humides du Cameroun,

Revue Scientifque et Technique Forêt et Environnement du Bassin du Congo 6:81-83.
Treseder KK (2013). The extent of mycorrhizal colonization of roots and its influence on plant growth and phosphorus content. Plant and Soil 371:1-13.
Trouvelot A, Kough JL, Gianinazzi-Pearson V (1986). Measurement of the VA mycorrhization rate of a root system. Research and estimation methods of functional significance. In: Physiological and genetic aspects of mycorrhizae, Dijon, 1985. INRA (ed.), pp. 217-221.
Van der Heijden MG, Boller T, Wiemken A, Sanders IR (1998). Different arbuscular mycorrhizal fungal species are potential determinants of plant community structure. Ecology 79(6):20822091.
Walder F, van der Heijden MGA (2015). Regulation of resource exchange in the arbuscular mycorrhizal symbiosis. Nature Plants 1:15159
Zhang F, Jia-Dong HE, Qiu-Dan NI, Qiang-Sheng WU, Zou YN (2018). Enhancement of drought tolerance in trifoliate orange by mycorrhiza: changes in root sucrose and proline metabolisms. Notulae Botanicae Horti Agrobotanici Cluj-Napoca 46:270.

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Evaluation of chemical and elemental constituents of *Centella asiatica* leaf meal

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Medicinal plants include various types of plants used in herbalism or herbal medicine. It is the use of plants for medicinal purposes, and the study of such uses. The use of medicinal plants is gaining popularity all over the world; hence the need to exploit various plant that could be of economic importance to both man and animal. *Centella asiatica* is one of such plants that are underutilized. Therefore, investigating the chemical and elemental constituents of *C. asiatica* leaf meal is very vital. The phytochemical analyses, proximate composition, vitamin and mineral concentrations were determined using standard procedures. Considerable quantity of phytochemical compounds such as phenolic, saponin, flavonoids, phytate and tannin were determined and the values obtained for these components were 2.75, 8.20, 12.85, 0.76 and 0.0%, respectively. While for the proximate: 95.76, 2.77, 12.40, 2.80, 2.40 and 75.44% were obtained for dry matter, crude fiber, crude protein, ash, ether extract and carbohydrate. The leaf meal contains appreciable quantity of calcium, magnesium, iron, phosphorus and sodium. Vitamins A, C and B_6 are readily available in the leaf meal while E and B_1 are not available. The nutrient composition of *C. asiatica* revealed that it contains some bioactive components which can serve as feed supplements in animal production and improve human health.

Key words: Chemical analysis, *Centella asiatica*, vitamins, minerals, additives, proximate.

INTRODUCTION

Plants have been used for medicinal purposes long before prehistoric period. These medicinal plants are also used as food, flavonoid, medicine or perfume and also in certain spiritual activities. Plants are used both by traditional herbalists and pharmacists for synthetic preparations in the pharmaceutical industries and for management and treatments of different diseases that affect man and animals (Tibi, 2012). The knowledge of medicinal plants has continued to be useful in the production of drugs, food, spice and feed additives. The research into plants bioactive substances has contributed immensely to the betterment of animal and human. Herbs and vegetables, especially leaves, are important sources of vitamins, minerals, fiber, and some essential amino acids.

Currently, as a result of resistance in the use of

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antibiotics in treating both human and animal diseases the use have been restricted in numerous countries in the world. Therefore, there is need for alternative means to address the issue (Laxminarayan et al., 2015). The use of herbs is gaining importance in animal production and human health due to harmful residual effects and cost effectiveness from antibiotics. Herbs, spices, essential oils, extract or oleoresins contain myriad highly active secondary plant metabolites unfolding a broad range of therapeutic effects. They can stimulate feed intake and endogenous secretions or have antimicrobial or anthelmintic activity. A major field of application of herbs is the protection of animals and their products against oxidation and improves human health. Plant such as: Ginger (*Zingiber officinale),* garlic (*Allium sativum*), cinnamon ((*Cinnamo mumzeylanicum*), thyme (*Thymus vulgaris*), Parsley (*Petroselinum crispum*) have been reported to increase digestion, increase appetite, antiseptic and gastric stimulant (Mirzaei-Aghsaghali, 2012)

Centella asiatica L. (Gotu Kola) Urban (*Gotu Kola coriacea* Nannfd., *Hydrocotyle asiatica* L., *Hydrocotyle lunata* Lam., and *Trisanthus cochinchinensis* Lour.) is a tropical medicinal plant from Apiaceae family native to Southeast Asian countries such as India, Sri Lanka, China, Indonesia, and Malaysia as well as South Africa and Madagascar (Jamil et al., 2007). It is native to the warmer regions of both hemispheres. This plant grows wild in damp, shady places up to 7000 ft. and can be commonly seen along banks of rivers, streams, ponds, and irrigated fields. It also grows along stone walls or other rocky areas at elevation of approximately 2000 ft. This plant is among the underutilized plant with few information on the chemical and proximate constituent therefore, to understand the roles played in human, animal nutrition and health, information on proximate, mineral, vitamin and phytochemical composition is crucial to the understanding of the mode of action of these medicinal plant in general. This will be useful for the nutritional and health education of the public as a means to improve their well-being.

MATERIALS AND METHODS

Leaf meal preparation

Fresh leaves of *Centella asiatica* were identified by an Agronomist from the Agronomy Unit of the Department of Agriculture, Babcock University, Ilishan Remo,Ogun State, Nigeria. The plant was harvested around the Teaching and Research Farm of the Department ofAgriculture and Industrial Technology, Babcock University, Ilishan-Remo, Ogun State, Nigerria. Harvesting was done between the hours of16:00 and 18:00 when the plant must have completed its lightstage of photosynthetic process for the day. The leaves were washed, chopped and air dried at an average room temperature of 27°C until when it's properly dried and pulverized with a blender to obtain a fine powder. The powdered sample was stored at 4°C in a dry, clean container with lid for further analysis.

Chemical analysis

Phytochemical analyses

Phytochemical analyses were conducted to determine the presence of phytate, saponin, flavanoid, tannin and alkaloid while the quantification of saponin was done by afrosimetric method (Koziol, 1991). The gravimetric method of Haborne (1973) was used in determination of alkaloid and flavonoid contents. All the analyses were done using triplicate samples.

Proximate analysis

The moisture content was determined by drying at 105°C in an oven until a constant weight was reached. For total ash determination, the leaf sample was weighed and converted to dry ash in a muffle furnace at 450°C and at 550°C for incineration. The crude fat content was determined by extraction with n-hexane, using a Soxhlet apparatus. All these determinations were carried out according to AOAC (1990). Kjeldahl method was used for crude protein determination. Carbohydrate content was determined by calculating the difference between the sums of all the proximate compositions from 100%.

Vitamin and minerals analysis

Vitamin analyses were carried out according to the method of Martin-Prevel et al. (1984). Mineral analyses were carried out using an atomic absorption spectrophotometer (AAS) calcium, magnesium and Iron. Atomic emission spectrophotometer technique was used to determine sodium content.

RESULTS

Phytochemicals

The analysis of *C. asiatica* in Table 1 revealed that they contained appreciable quantity of phytochemicals. Phytochemical composition of *C. asiatica* leaf contains 2.75% phenolic, saponin 8.2%, flavonoids 12.85%, phytate 0.76% and 0.00% tannin.

Proximate

The proximate composition as contained in Table 2 revealed the dry matter to be 95.76, 2.77 12.40% crude protein, 2.80 and 2.40% crude fiber and 75.44% carbohydrate. Value of dry matter obtained in this study indicated that the leaf meal contains little quantity of moisture content.

Vitamin

Vitamin composition of *C. asiatica* leaf meal (CALM) indicated that it contains some vitamins which could be of

Table 1. Phytochemical composition of *Centella asiatica* leaf meal.

Parameter	Composition	Means±Std
Phenolic	2.75	2.75±0.05
Saponin	8.20	8.20±0.40
Flavonoids	12.85	12.85±0.05
Tannin	0.00	0.00±0.00
Alkaloid	2.90	2.90±0.30
Phytate	0.76	0.76±0.04

Table 2. Proximate composition of *C. asiatica* leaf meal.

Parameter (%)	Composition	Means±Std
Dry matter	95.76	95.76±0.01
Crude fibre	2.77	2.77±0.010
Crude protein	12.40	12.40±.0.10
Ash	2.80	2.80±0.10
Crude fat	2.40	2.40±0.10
Carbohydrate	75.44	75.44±0.01

Table 3. Vitamin composition of *C. asiatica* leaf meal.

Vitamins mg/100 g	Composition	Mean±std
A	0.39	0.39 ± 0.01
E	N/A	-
C	0.76	0.76 ± 0.01
B1	N/A	-
B6	0.78	0.78 ± 0.01

NA: not available.

Table 4. Minerals composition of *C. asiatica* leaf meal.

Minerals g/100 g	Composition	Mean±std
Calcium	24.38	24.38±0.3
Magnesium	3.18	3.14 ±0.05
Iron	0.20	0.20±0.1
Phosphorus	3.14	3.14±0.01
Sodium	8.20	8.20±0.06

importance to man. 0.39 mg/100 g was obtained for vitamin A, no trace of vitamin E and B_1, 0.76 mg/100 g of vitamin C and 0.78 mg/100 g of vitamin B_6 (Table 3).

Minerals

Appreciable quantity of minerals was observed in the *C. asiatica* leaf, it contains 24.38 g/100 g of calcium, 3.18 g/100 g magnesium, 0.20 g/100 g iron, 3.14 and 8.20 g/100 g sodium (Table 4). The quantity of calcium in CALM could be a good source of food supplement to both man and human suffering from calcium deficiency.

DISCUSSION

Phytochemical analyses of *C. asiatica* leaf meal (CALM) in Table 1 revealed the presence of tannin, flavonoids, saponin, alkaloid, phytate and phenolic compounds. The phytochemical constituents isolated from CALM have been reported to have hypotensive, anti-inflammatory, antioxidant, antifungal, antimicrobial and antibacterial activities (Wadood et al., 2013).

Biological activities of phenolic acids include increase in secretion of bile; reduce blood cholesterol and lipid levels. Okwu and Vitus (2008) also revealed that phenol modifies the prostaglandin pathways, thereby protecting platelet from clumping. Value obtained for phenolic compound in this study was 2.75%. However, Okwu and Vitus (2008) obtain a lower value of 0.75 and 0.09% from back stem and leaves of *Mangifera indica*. Higher value of phenol obtained in this study could be of health benefit to both man and animal when consumed.

The flavonoids obtained in this study was appreciable12.85%; flavonoids have been reported to exert multiple biological properties including anti-microbial, cytotoxicity, anti-inflammatory and antitumor activities. They act as powerful antioxidants which can protect the human body from free radicals and reactive oxygen species (Nakatani, 2000; Wei and Shibamoto, 2007; Dharmendra and Abhislick, 2013; Gupta et al., 2013). Flavonoids constitute a wide range of substances that play important role in protecting biological systems against the harmful effects of oxidative processes on macromolecules such as carbohydrates, proteins, lipids, and DNA (Atmani et al., 2009). The presence of flavonoids in *Centella asiatica* supports the findings of Das (2011) who observes flavonoids derivatives in *C. asiatica* leaf. Roy (2018) also reports the presence of flavonoids derivatives in *C. asiatica* leaf meal. Flavonoids could prevent oxidative reactions and improve health status of both plant and animal. Tannin has been reported to form complexes with dietary protein, thereby inhibiting protein metabolism and utilization in monogastric animals (Vaithiyanathan et al., 1993). Tannin strongly inhibits digestive enzymes (Kumar, 1992). Absence of tannin in *C. asiatica* indicated that protein metabolism, feed utilization and mineral absorption will not be hampered if consumed. The value obtained in this study was 0.00% which was lower to the value of 3.03% obtained by Alagbe (2019) in *C. asiatica* leaf this might be due to difference in location, age of harvest, morphotypes of plant used for the study or extraction method. Marrippan (2018) reports the absence of tannin in *C. asiatica* when hexane and chloroform was used as extraction media.

Suma et al. (2014) report that zinc and iron deficiency symptoms have occurred in man and poultry birds when fed diets high in phytic acid. Phytate obtained in this study was 0.76%. This value was similar to the value of 0.77% by Alagbe (2019) for *C. asiatica* leaf meal. Olumide et al. (2019) also reported a lower values of 0.03, 0.17 and 0.02% respectively for *Ocinum gratissimum, Vernonia amygdalina* and *Moringa oleifera,* the higher value obtained in this study indicated that *C. asiatica* could be effectively utilized when moderately incorporated into animal feed while cautions as per moderation must be taken for human consumption considering the relatively high content of phytic acid in *C. asiatica*.

Alkaloids are essential for protecting and ensuring the survival of plant because they ensure their survival against micro-organisms (antibacterial and antifungal activities). Alkaloids contained plants can be used as spices and drugs. Alkaloids that have stimulant property as caffeine, nicotine and morphine are used as analgesic and quinine as antimalarial drug (Saxena et al., 2013). Pharmacological activities of alkaloids include antihypertensive, anticancer and antiarrhythmic effect. The value of alkaloids obtained in this study (2.90%) support the findings of Roy (2018) in quantitative analysis of *C. asiatica* leaf extract. Alagbe (2019) also obtained a lower value of 2.03% for *C. asiatica* leaf. The presence of alkaloids in this plant could serve as protective role in animal and as constituent of most valuable drugs.

Akindahunsi and Salawu (2005) noted that saponin though non-toxicsaponinexhibits cytotoxic effects and growth inhibition against a variety of cells making it to have anti-inflammatory and anticancer properties. AICR (1997) noted that saponin showed tumor inhibiting activity in animals. The value obtained for saponin in this study was 8.20% which could be of health benefit to both human and animal.

The proximate composition of *C. asiatica* leaf meal in Table 2 indicated that dry matter content of *C. asiatica* was 95.76%, which was higher than the value obtain by Alagbe (2019) who reported 90.44% for *C. asiatica* leaf. Nworgu et al. (2007) reported a lower value of 87.96% for fluted pumpkin and Abu et al. (2015) observed lower value of 92.40% for cassava leaf. Ibironke et al. (2013) obtained 48.09% for *Sphenocentrum jollyanum* plant. This might be as a result of level of dryness of the leaf, method of drying and age at harvest.

The percentage crude protein obtained in this study was 12.40% which was similar to 13.06% obtained by Alagbe (2019) in C. *asiatica* leaf. Oku (2018) obtained 16.80% in *Ipomea involucrate* leaf. Discrepancy observed might be due to plant morphotypes and stage of harvest. The value obtained in this study is an indication that *C. asiatica* might be a good source of dietary protein supplement.

Carbohydrate obtained in this study was 75.44%. However, Olumide *et al*. (2019) report low value of 49.75, 44.90 and 31.70% for *O. gratissimum*, *V. amygdalina* and *M. oleifera.* CALM could be a potential energy sourceto human and animal.

Crude fibre obtained in this study was 2.77% which is lower than the value, 24.8, 11.40,12.00%, 12.93 and 5.60 observe for *Microdesmispuberula*, cassava leaf meal, neem leaf meal, mucuna leaf meal, and pawpaw leaf meal (Esonu et al., 2002; Iheukwumere et al., 2008; Onyimonyi et al., 2009; Emenalom et al., 2009) respectively. The relatively low crude fibre of CALM makes it a potentially good feed stuff for poultry production.

Ash content obtained in CALM in this study was 2.80% which was lower than the value of 4.07 and 8.45% obtain by Alagbe (2019) and Onyimonyi et al. (2009) in *C. asiatica leaf* and pawpaw leaf respectively and higher than the range of 0.38 to 1.9% for selected vegetables grown in Peshawar (Bangash et al., 2011).

Quantity of ash obtained support the claim of Das (2011), Hashim (2011) and Josh et al*.* (2013) that *C. asiatica* leaves contains minerals such as calcium, magnesium, iron, sodium and phosphorus.

Table 3 showed vitamin composition of *C. asiatica* leaf meal, the value of Vitamin A in this study was 0.39 mg/100 g, this result was similar to the report of Hashim (2011) who obtained 0.44 mg/100 g of Vitamin A in *C. asiatica* leaf, Josh et al. (2013) observe 0.0 mg/gvitamin A in *C. asiatica*leave*.* The variation observed in vitamin composition of CALM could be due to age of harvest ofplant, drying methods, plant morphotypes or environmental factors. The different varieties of *C*. *asiatica* could be exploited as a good source of pro-vitamin A and lutein to overcome vitamin A deficiency as well as age-related muscular degeneration (Chandrika et al., 2006).

Vitamin C obtained in this study (0.76 mg/100 g) was lower compared to the values, 11 mg/100 g and 9.73 mg/g by Josh et al*.* (2013) and Das (2011) respectively. Edelman and Colt (2016) obtain 2 mg/100 g in lentil, considerable higher value is obtained from kale, spinach and duck weed. The lower value obtained in this study might be due to difference in leaf composition and dehydration process, as ascorbic acid show highest reduction with the dehydration processes (Gupta et al., 2013). Vitamins E and B_1 were not detected, this connote that CALM is not a good source of vitamin E and B_1.

The analysis indicated that CALM contains vitamin B_6 also known as pyridoxine a water soluble vitamin that the body needs for several functions. It is significant to protein, fat and carbohydrate metabolism and the creation of red blood cells and neurotransmitters. Vitamin B_6 is used in prevention and treatment of anemia caused by deficiency, it prevent clogged arteries and reduce heart disease risk (Lizzie, 2018). CALM could serve as good supplement of vitamin B_6.

Mineral composition of CALM in Table 4 indicated that it contains appreciable amount of minerals. The value obtained for calcium was 24.38 g/100 g. This was higher compared to the 2.10 g/100 g obtained by Chandrika et al. (2011) in different varieties of *C. asiatica* examined. Alagbe (2019) report a lower value of 10.20 mg/100 g for *C. asiatica* leaf meal (CALM). Edelman and Colt (2016) also obtain lower value of 0.34, 8.46, 10.36 and 6.00 mg/100g in lentil, kale, spinach and duckweed while Okwu and Vitus (2008) report 1.41, 3.82 mg/100 g for mango stem and mango leaves which are also used as phytogenic plants. The variations obtained in different varieties of CALM might be due to plant morphotype, method of extraction, storage time and age atharvest. This finding support the claim of Chandrika et al. (2011) that CALM can be used as non-expensive calcium feed supplement.

Magnesium is often used as a laxative. The value obtained for magnesium in this study was 3.18 g/100 g, this value was higher than the values of 0.2g/100g and 0.4g/100g obtain by Chandrika et al. (2011) for different varieties of CALM. However, Alagbe (2019) report a higher value of 9.06 (mg/100 g) for *C. asiatica* whereas, Okwu et al. (2008) report a lower value of 0.46 and 0.91 mg/100 g for mango stem bark and leave.

Iron constituent in this study was 0.20 (mg/100g), this was lower than the value obtain by Chandrika et al (2011) who obtained 0.40, 0.50, and 0.29 mg/100g respectively from different varieties of *C. asiatica*.

The composition of sodium was 8.20 g/100 g; this was higher than the value of 2.2 g/100 g, 1.1 g/100 g and 2.6 g/100 g obtain in *C. asiatica* by Chandrika et al. *(*2011) and higher than value 0.97 mg/100 g reported by Gupta (2004) in *C. asiatica*. The value obtained in this study was also higher than the value obtain in *Hymenocardia ulmoides* and *V. ferruginea* leaves by Andzouana and Mombouli (2011).

Phosphorus component was 3.14 g/100 g which could help in appetite control and improve feed utilization, maintain blood sugar levels and normal heart contraction (Linder, 1991). The value obtained in this study is higher than the value 0.16 mg/100 g by Ngozi et al. (2017).

According to different studies done with CALM in the previous decade, the nutrients content shows relatively close values but in some instances, big variations are also seen (Das, 2011; Hashim, 2011; Joshi and Chaturvedi, 2013). These values may vary considerably depending on the analytical method, biotic and abiotic factors. The presence of considerable quantity of mineral component in CALM could be of importance in reducing anemia, proper functioning of the nervous system and carbohydrate metabolism.

Conclusion

It can be concluded from the study that *C. asiatica* leaf meal could serve as a good source of vitamins and mineral supplements to human and animal. The leaves also contain phytochemical components such as flavonoids, alkaloids which are of good health benefit to both human and animal when consume for therapeutic purpose and could be of economic importance to the pharmaceutical industry.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Abu OA, Olaleru IF, Oke TD, Adepegba VA, Usman B (2015). Performance ofbroiler chicken fed dietscontainingcassava peel and leaf meals asreplacements for maize and soya bean meal. International Journal of Science and Technology 4:169-172.

Association of Official Analytical Chemists (AOAC) (1990). Official Methods of Analysis. 15th Edn. AOAC, Washington, DC. USA, pp. 200-210.

AICR (1997). American Institute for Cancer Research. Food, Nutrition and the Prevention of Cancer: A global Perspective, P. 423.

Alagbe JO (2019). Effects of Dried Centella Asiatica Leaf Meal as a Herbal Feed Additive on the Growth Performance, Heamatology And Serum Biochemistry of Broiler Chicken. International Journal of Animal Research 3:23

Andzouana M, Mombouli JB (2011). Chemical composition and phytochemical screening of the leaves of *Hymenocardiaulmoides* and *Vitexferruginea*. Pakistan Journal of Nutrition 10:1183-1189.

Akindahunsi AA, Salawu SO (2005). Phytochemical screening, nutrient and antinutrient composition of selected tropical green leafy vegetables. African Journal of Biotechnology 4(6):497-501.

Bangash JA, Arif M, Khan F, Amin-Ur-Rahman, Hussain I (2011).Proximate composition, minerals and vitamins content of selected vegetables grown in Peshawar. Journal of the Chemical Society of Pakistan 33(1):118-122.

Chandrika UG, Svanberg U, A'Jansz ER (2006). *In vitro* accessibility of Pcarotene from cooked Sri Lankan green leafy vegetables and their estimatedcontribution to vitamin A requirement. Journal of the Science of Food and Agriculture 86:54-61.

Chandrika UG, Salim N, Wijepala GD, Perera, KSU, Goonetilleke AKE (2011). Carotenoid and mineral content of different morphotypes of *Centella asiatica* L. (Gotu kola). International Journal of Food Sciences and Nutrition 62:552-557.

Das AJ (2011). Review on nutritional, medicinal and pharmacological properties of Centella asiatica (Indian pennywort). Journal of Biologically Active Products from Nature 4:216-228.

Dharmendra S, Abhislick G (2013). Phytochemistry of medicinal plants. Journal of Pharmacognosy and Phytochemistry pp. 168-182.

Edelman M, Colt M (2016). Nutrient value of leaf vs. seed. Frontiers in Chemistry 4:1-32.

Emenalom OO, Esonu BO, Etuk EB, Anaba C (2009). Effect of *Mucunapruriens* (Velvet Bean) leaf meal on Performance and blood composition of finisher broiler chickens. Nigerian Journal of Animal Production 36:52-60.

Esonu BO, Iheukwumere FC, OO. Emenalom Uchegbu MC, Etuk EB (2002). Performance, nutrient utilisation and organ characteristics of broilers fed *Microdesmis puberula* leaf meal. Retrieved from https://www.lrrd.cipav.org.co/lrrd14/6/eson146.htm

Gupta S, Gown, BS, Lakshmi AJ, Prakash J (2013). Retention of nutrients ingreen leafy vegetables on dehydration. The Journal of Food Science and Technology 50(5):918-925.

Hashi, P (2011). *Centella asiatica* in food and beverage applications and its potential antioxidant and neuroprotective effect. International Food Research Journal 18(4):1215-1222.

Harborne JB (1973). Phytochemical methods: A guide to modern techniques of plant analysis. Chapman and Hall Ltd, London P. 279.

Joshi K, Chaturvedi P (2013). Therapeutic efficiency of *Centellaasiatica* (1.) urbAn underutilized green leafyvegetable: An overview. International Journal of Pharma and Bio Sciences 4:135-149.

Iheukwumere FC, Ndubuisi EC, Mazi EA Onyekwere MU (2008). Performance, nutrient utilization and organ characteristics of broilers fed cassava leaf meal (*Manihot esculenta* Crantz). Pakistan Journal of Nutrition 7:13-16.

Koziol MJ (1991). Afrosimetric estimation of threshold saponin concentration forbitterness in quinoa (*Chenopodium quinoa* Willd). Journal of the Science of Food and Agriculture 54(2):211-219.

Kumar R (1992). *Prosopis cineraria* leaf tannins. Their inhibiting effect upon ruminal cellulose and the recovery of inhibition by polyethylene glycol-4000. In: Plantpolyphenols, synthesis, Properties and significance. Basic Life Science 59:699-704.

Laxminarayan R, Boeckel T, Teillant A (2015).The Economic Costs ofWithdrawing Antimicrobial Growth promoters from Laxminarayan the Livestock Sector OECDFOOD, Agriculture and fisheries paper n°78 © oecd.

Linder MC (1991) nutritional biochemistry and metabolism with clinical applications. Appeleton and lange Norwalk 2:191-212

Mirzaei-Aghsaghali A (2012). Importance of medical herbs in animal feeding: A review. Annals of Biological Research 3(2):918-923.

Martin-Prevel P, Gagnard J, Dautier P (1984). Dans le Controle de L alimentation des Plantes Temperees et Tropicales. Technique et Documentation, Paris. Journal of Cell Biology 133:683-693.

Mariappan S (2018). Preliminary phytochemical screening of bioactive compounds from leaves of *Centella asiatica* L. Urban. International Journal of Biology Research 3(1):57-65.

Ngozi KA, Chimaroke O, Chima A, Ekeleme E, Jennifer CO (2017). Phytochemical, Proximate Analysis, Vitamin and Mineral Composition of Aqueous Extract of Ficus capensis leaves in South Eastern Nigeria. Journal of Applied Pharmaceutical Science 7(03):117-122.

Nakatani N (2000). Phenolic antioxidants from herbs and spices. Biofactors 13:141-146.

Nworgu FC, Ekemezie AA, Ladele AO, Akinrolabu BM (2007). Performance of broiler Chickens served heattreated flutedpumpkin (*Telfariaoccidentalis*)leaves extract supplement. African Journal of Biotechnology 6(6):818-825,

Okwu E, Vitus E (2008). Evaluation of the phytochemical composition of mango (*Mangiferaindicalinn*) stem bark. International Journal of Chemical Science 6(2):705-716.

Oku ED, Anwan AH, Udoma EE (2018). Comparative Phytochemical, Proximate, Vitamins and Mineral Nutrient Composition of Leaf, Stem, and Root of *Ipomea involucrata* and *Milletia aboensis* from Southern Nigeria. Saudi Journal of Medical and Pharmaceutical Sciences pp. 1-6.

Olumide MD, Akinola AS (2018). Effect of scent leaf meal (ocimumgratissium)supplementation on performancecarcass and meat quality of broiler chicken. Nigerian Journal of Animal Production 45(3):228-236

Olumide MD, Ajayi OA, Akinboye OE (2019). Comparative study of proximate, mineral and phytochemical analysis of the leaves of *Ocimum gratissimum*, *Vernonia amygdalina* and *Moringa oleifera.* Journal of Medicinal Plants Research 13(15):351-356.

Onyimonyi AE, Adeyemi O, Okeke GC (2009). Performance and Economic characteristics of broilers fedvarying dietary levels of Neem leaf meal (*Azadirachtaindica*). International Journal of Poultry Science 8:256-259

Tibi B (2012). Islamic Humanism vs. Islamism:Cross-Civilizational Bridging. Soundings: An Interdisciplinary Journal, 95(3):230-254.

Wei A, Shibamoto T (2007). Antioxidant activities and volatile constituents of various essential oils. Journal of Agricultural and Food Chemistry 55:1737-1742.

Saxena M, Saxena J, Nema R, Dharmendra S, Gupta (2013). A Phytochemistry of Medicinal Plants. Journal of Pharmacognosy and Phytochemistry Online Available at www.phytojournal.com

Suma PF, Urooj A (2014). Nutrients, antinutrients&bioaccessible mineral content (invitro) of pearl millet as influenced by milling. Journal of Food Science and Technology 51(4):756-761.

Roy AR (2018). Qualitative and Quantitative Phytochemical Analysis of Centellaasiatica. Natural Products Chemistry and Research 6:4

Wadood A, Ghufran M, Jamal SB, Naeem M, Khan A (2013). Phytochemical Analysis of Medicinal Plants Occurring in Local Area of Mardan. Biochemistry and Analytical Biochemistry 2:144. doi: 10.4172/2161-1009.1000144

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Technical efficiency in tomato production among smallholder farmers in Kirinyaga County, Kenya

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The study was conducted to assess tomato productivity and determine characteristics that influence technical efficiency among smallholder farmers in Kirinyaga County using the production function approach. Data were collected by administering structured questionnaires to a sample of 384 respondents randomly selected from six wards using multistage stratified and probability proportionate to size sampling procedures. The study adopted cross-sectional survey design and primary data on tomato yield, production system, input usage and farmer demographics were collected. The stochastic Cobb Douglas production function was used to estimate the frontier production and efficiency levels using maximum likelihood. Tobit multiple regression was used to determine farm and farmer characteristics that impact technical efficiency. Results showed that respondents were inefficient with an average technical efficiency of 39.55% with greenhouse more efficient than open field system. Household size, production systems, seed type, fertilizer, extension and market information significantly and positively influenced technical efficiency, while land size was significant and inversely influenced technical efficiency. Results revealed a possibility to increase technical efficiency in tomato production using certified seeds and recommended fertilizer levels. In addition, policy interventions aimed at subsidizing costs of establishing greenhouses would serve as an incentive to motivate farmers to use technologies in tomato production.

Key words: Cobb Douglas production function, production systems**,** technical efficiency, Tobit regression

INTRODUCTION

Comparable to other Sub-Saharan African countries, Kenya continues to rely on agriculture for food and economic development (Ochilo et al., 2019). The sector is a key economic pillar contributing 24% of the gross domestic product and about 65% of exports (Nyamwamu, 2016). Smallholder farmers dominate the sector with farms ranging from 0.2 to 3 hectares and produces over 70% of total agricultural output (Ndirangu et al., 2018). Horticulture forms the bulk of agriculture with vegetables accounting for 80% of growers and 60% of exports (Yabs and Awuor, 2016). Tomato is among the widely cultivated vegetables and is ranked second after potato in terms of production and value (Mitra and Yunus, 2018). In recent past, the Kenyan government has devised mechanisms to improve productivity among smallholder farmers (Wambua et al., 2019). In tomato

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production, this involved development of disease resistant varieties, quality fertilizers, effective pesticides and technologies aimed at reducing production costs (GoK, 2018).

Despite these efforts, the country has not attained food sufficiency like is the case in other developing countries. To overcome this, it is necessary to increase agricultural yield by improving production efficiency.

Tomato is among vegetables mainly grown in open fields and greenhouse production systems (Nyamwamu, 2016). The crop is extensively cultivated, accounting for about 7% of horticulture and 14% of vegetable production (Mwangi et al., 2015). On average, the country records 410,033 tons of tomato with area under cultivation escalating from 18,178 ha to 20,111 ha between 2011 and 2016. (Mitra and Yunus, 2018).This places Kenya among top tomato producers in Sub Saharan Africa (Ochilo et al., 2019). In Kenya, tomatoes are grown in areas with altitudes that range from 1150 and 1800m above sea level (Mwangi et al., 2015). Mwangi et al. (2015) also noted that Kirinyaga County leads (14%), in tomato production followed by Kajiado (9%) and Taita Taveta (7%). The crop contributes to income generation, foreign exchange, poverty alleviation, and employment creation especially to rural populations (Singh et al., 2017). In spite of this substantial contribution to rural economies in the region, tomato production encounters challenges of low productivity (Geoffrey et al., 2014). Chepng'etich et al. (2015) explains that actual yields remain below the maximum attainable levels with Sub Saharan Africa recording an agricultural produce that is below the global average.

In Kenya, despite efforts to improve tomato production by introducing modern technologies such as greenhouses, productivity declined from 22.4 tons in 2011 to 17.9 tons in 2015 and 16.9 tons in 2016 (Tabe and Molua, 2017). Ochilo et al. (2019) noted that deviations persisted in 2018 with an average yield of 12 tons/ha against a potential yield of 30.7 tons per ha. This was below an average of 35 tons/ha in Egypt and 120 tons/ha in France (Najjuma et al., 2016).

The low productivity in agriculture has resulted from the inability of farmers to fully utilize available technologies hence leading to inefficiencies in production (Kumar et al., 2018). Further, Wanjiku (2015) indicated that land available for agricultural production has reduced due to the enormous population growth, extensive soil degradation and intensified land fragmentation thus lowering productivity. Besides this, the high poverty levels entwined with the limitation of factors of production has made it difficult for farmers to increase production through use of more resources (Simwaka et al., 2013). The existence of inefficiencies in agricultural production implies the need to examine technical efficiency of agricultural production particularly among smallholder tomato farmers (Wahid et al., 2017). Kumar et al. (2018) explained that an understanding on levels of technical efficiency can be valuable in solving the problem of low productivity in agriculture.

Ochilo et al. (2019) indicated that production and productivity in agriculture can be improved through increased input use and increasing technical efficiency levels of producers. Measuring technical efficiency helps compare the performance of farmers and identify factors that explain inefficiencies (Kassa and Demissie, 2019). Technical efficiency depicts the producer's ability to achieve optimal production from the available resources and level of technology (Shettima et al., 2015). Dessale (2019) explained that the performance of a producer and factors that affect production are important aspects in quantifying technical efficiency. Technically, efficient farms produce along the frontier while inefficient producers lie below the frontier production function (Tirra et al., 2019). In addition, farms closer to the production frontier are more technically efficient than those far from the frontier (Katungwe et al., 2017). Improving efficiency enables farmers increase yields without additional inputs and technologies thus enhanced productivity (Saavedra et al., 2017). Among smallholder farmers, inefficiencies may also arise from farm and farmer characteristics (Singh et al., 2017).

The Kenyan tomato industry is set to improve given the governments' pursuit to realize the Big Four Agenda which targets to achieve food security by boosting productivity among smallholder farmers (Tirra et al., 2019). Despite the significance of increasing productivity, literature on technical efficiency a key aspect in increasing agricultural production is scanty. In addition, very few studies have profiled and compared technical efficiency of tomato production between open field and greenhouse production systems in Kenya particularly Kirinyaga County. This is so despite the County leading in tomato production in Kenya. In addition, the component of describing tomato farmers based on production systems and determining their technical efficiency is limited. Besides this, it is necessary to investigate the causes of technical inefficiency and low productivity among the smallholder tomato farmers.

The main goal of this study was therefore to examine the level of technical efficiency of smallholder tomato farmers in Kirinyaga County and identify characteristics that influence technical efficiency. The study conducted an in depth analysis of technical efficiency by determining the frontier production function and the yield gap of tomato farmers from the maximum achievable output. This was computed given the existing technology and level of inputs by maximizing output per unit of input. This highlighted the extent to which factors of production such as seeds, fertilizers and pesticides account for variations in yields. The results revealed that technical efficiency for tomato farmers in Kirinyaga remained low with greenhouse farmers more efficient than open field farmers.

The study provides relevance in practical and theoretical setups. At the practical level, measuring technical efficiency and identifying factors that influence it among smallholder farmers provides meaningful information to policy makers in the formulation of strategies that are likely to improve the producer technical efficiency. From the microeconomic view, the identification of factors that improve farm performance is of utmost importance and through utilization of research based information from such studies farm efficiency may increase thus better returns. At the theoretical level, the study aims at contributing to existing literature and understanding of producer technical performance in rural areas of developing countries.

MATERIALS AND METHODS

Description of study area

The study was conducted in Kirinyaga central and Mwea west sub-counties of Kirinyaga County which are the major tomato growing zones in the County and are located in the lowland and midland agro-ecological zones (GoK, 2018). The County is located along the slopes of Mt. Kenya and lies between latitudes 0°1' and 0°40', and latitudes 37° and 38° East with altitude between 1158 and 5199 metres above sea level (Mwangi et al., 2015). Rainfall in the study area is bimodal with long rains occurring from March to May and short rains occurring from October to December with quantities ranging from 1,212 to 2,146 mm (GoK, 2018). On average, temperatures range from 8.1°C to 30.3°C. Agriculture is the major economic activity with majority (70%) of farmers being smallholders (MoA, 2011). Tomato is among the promising horticultural crops.

Sample size

The sample size of this study was 384 smallholder tomato farmers who were obtained from major tomato producing areas in Kirinyaga County. The following formula was used to determine the sample size as applied by Narcisse (2017).

$$n_o = \frac{z^2\ pq}{d^2} \quad (1)$$

Where n_o is the desired sample size, z is the standard normal value (1.96), p is the proportion of households producing tomatoes in small scale in Kirinyaga County, q equals 1-p and d is the desired precision level or level of significance (5%). The study adopted a proportion of 50% that the respondents possess the characteristic being measured.

Research design and sampling technique

The study employed a cross-sectional survey research design which ensures accuracy (Bhattaraiet al., 2016) and estimates the extent of realizing sample outcome for a given population (Okonya and Kroschel, 2015). The study embraced multistage stratified random sampling to sample respondents to be interviewed. The two sub counties were purposively selected and from each sub county, wards were selected based on the concentration of tomato production. Six wards that mainly grow tomatoes in small-scale were considered. Since the expected number of greenhouse farmers was low, a census survey was conducted and a total of 78 greenhouse farmers obtained. Consequently, from each ward, probability proportionate to size sampling technique was applied using the sampling frame to select 306 open field farmers. The number of farmers from each ward was determined using the following formula as applied by Wambua et al. (2019).

$$K = \frac{P}{M} * 306 \quad (2)$$

Where, k = number of farmers to be interviewed from each ward; P= the number of smallholder tomato farmers in each selected ward and M= total number of smallholder tomato farmers in the selected wards.

Data analysis

The study collected both qualitative and quantitative data. The Statistical Packages for Social Sciences (SPSS) was used for descriptive analysis. The stochastic frontier production function and the censored Tobit regression model in STATA version 13.0 were used as econometric models. Means, percentages and standard deviations were used to describe the distribution of technical efficiency scores, socioeconomic characteristics of the respondents and the farm characteristics. The maximum likelihood estimation procedures were used to estimate the stochastic production function of the Cobb Douglas functional form. The estimated efficiency scores were regressed against the selected farm and farmer characteristics using Tobit multiple regression model to identify factors that influence technical efficiency.

Theoretical framework

The economic theory of production provides a methodical background for most studies on efficiency and productivity (Katungwe et al., 2017). Efficiency is regarded as the relative performance of transforming inputs into outputs. Agricultural productivity can be defined as the ratio of the value of total farm output to the value of the total inputs used in the farm production (Tabe and Molua, 2017). The economic theory gives a distinction between two forms of efficiency, allocative and technical efficiency and a farm that meets both is said to be economically efficient (Ndirangu et al., 2018). Frontier measures of efficiency imply that efficient farms operate along the production frontier. In addition, the amount by which a farm deviates from its frontier production is regarded as a measure of inefficiency (Chepng'etich et al., 2015). Tabe and Molua (2017) noted that research has categorized measures of efficiency into non-parametric and parametric approaches. The most common non parametric approach is the data envelopment analysis (DEA) which does not separate deviations in output into inefficiency and random errors. Further, it does not allow hypothesis testing for the fitness of the model (Parman and Featherstone, 2019). The stochastic frontier production is the frequently used parametric approach. This approach entails the description of the technology which may be restrictive in most cases (Ajibefun, 2008). Further, the stochastic frontier model attributes deviations from the frontier function into inefficiencies and random errors thus accurate and less sensitive to measurement errors in data (Ndirangu et al., 2018). In addition, it allows testing of hypothesis regarding the goodness of fit for the model. This model estimates the production function by fitting observed data and minimizing measures of their distance from the expected frontier

(Abdul and Isgin, 2016).The stochastic production frontier was initially proposed by Aigner et al. (1977) and Mueesen and Broeck (1977). The model is generally expressed as:

$$Y_i = f(X_i;\ \beta)\exp(V_i - U_i) \quad (3)$$

Where, 1 = 1, 2, 3… n, X_i is vector of input quantities used by the ith farmer, Y_i is tomato output of the ith farmer, β is a vector of parameters to be estimated. V_i is the distinctive error term that arises from measurement errors in input use and yield. U_i is a non-negative ($U_i \geq 0$) random variable with half normal distribution. It measures technical efficiency relative to the frontier production function (Battese and Coelli, 1995).

The computation of technical efficiency of an individual farm was achieved by comparing actual yield to optimal outputs. Technical efficiency of an individual farmer was defined as the ratio of actual output to the optimal production, constrained by the input levels used as shown below:

$$TE_i = \frac{Y_i}{Y_i^*} = \frac{[F(B,X) + (v_i + u_i)]}{[(B,X) + V_i)]}$$

Where, TE_i is the technical efficiency of an individual farmer, Y_i represents actual output and Y_i^* represents optimal output.

Empirical model

In this study, technical efficiency among smallholder tomato farmers in Kirinyaga County was measured using the stochastic frontier production which was based on the Cobb Douglas production function. Despite the limitation of the Cobb Douglas production functional form, it provides suitable representation of any production technology used. Further, it is capable of holding multiple input modelling and efficient in managing multicollinearity, heteroscedasticity, and correlation (Mitra and Yunus, 2018). The Cobb Douglas stochastic production frontier function is specified as:

$$\operatorname{Ln} Y_i = \beta_0 + \beta_1 \ln X_1 + \beta_2 \ln X_2 + \beta_3 \ln X_3 + \beta_4 \ln X_4 + \beta_5 \ln X_5 + V_i - U_i \quad (5)$$

Where, X_1 is seed quantity (g), X_2 is labour used (Mds/ha), X_3 is pesticides (L/ha), X_4 is fertilizer quantity (Kg/ha), X_5 is farm size (ha).

The Tobit censored regression model was used to investigate characteristics that affect technical efficiency among smallholder tomato farmers in Kirinyaga County. Since efficiency ranges from a minimum of 0 to a maximum of 1, the model was appropriate as it is a limited dependent variable model (Chepng'etich et al., 2015). The model, based on Battese and Coelli (1995) and applied by Tabe and Molua (2017) is specified as:

$$U_i = \alpha_0 + \alpha_1 X_1 + \alpha_2 X_2 + \alpha_3 X_3 + \alpha_4 X_4 + \cdots + \alpha_{17} X_{17} \quad (6)$$

Where,U_i= Technical efficiency scores, X_1 =Age, X_2 =Gender, X_3 =Household size, X_4 =Farmer group membership, X_5 = Experience in tomato production, X_6= Education, X_7= type of production system, X_8=Land tenure, X_9 =Seed type, X_{10}= Off farm income, X_{11}= Farm income, X_{12} =Land size under tomatoes, X_{13} = Fertilizer quantity, X_{14} = Access to extension services, X_{15}= Distance to market, X_{16} = Access to market information, X_{17} =Credit value.

RESULTS AND DISCUSSION

Continuous farm and farmer characteristics

The research conducted a descriptive analysis of the farm and farmer characteristics of the smallholder tomato producers in Kirinyaga County. The respondents used both open field and greenhouse production systems. Table 1 shows a comparison of the continuous factors of the respondents using open field and greenhouse production systems. In this study, open field system was considered as a conventional method of commercial tomato production in the open-air space without any protection from the environment. Further, the greenhouse system was conceptualized as growing tomatoes under a structure covered with transparent materials that transmit light for the growth of the plants as explained by Wachira et al. (2014).

From Table 1, the average age of the respondents was 37.03 years for the sample and ranged from 25 to 75 years. The results also revealed a mean of 36.36 and 39.64 years for open field and greenhouse farmers, respectively. The age differences between farmers in the two production systems were different at 1% level implying that greenhouse farmers were significantly advanced in age than their open field farmers. This shows that as tomato farmers progressed in age, they were more receptive to modern technologies of production.

Besides, the respondents apportioned an average credit of Ksh 29,930 for tomato production with Ksh 9,998 for open field and Ksh 108,121 for greenhouse farmers. Credit availability showed that farmers were empowered to timely procure improved inputs and adopt modern technologies. Differences in value of credited used in tomato production between the two production systems differed significantly at 1%. The large proportion used by the greenhouse farmers was informed by the high initial costs of investments required in this system while establishing the structures.

On average, the household size was 5.14 members for the sample, 5.16 members for open field and 5.08 members for greenhouse farmers. However, there were no statistical differences in household sizes between the two tomato production systems. In addition, the respondents recorded an average experience of 9.06 years in tomato farming. The mean number of years in tomato farming for open field system was 10.55 years and 3.26 years for the greenhouse farmers and the differences were significant. This suggests that open field farmers were significantly more experienced thus had more knowledge and understanding of tomato production than the greenhouse farmers. Regarding years spent while schooling by the head of household, open field smallholder farmers had a mean of 8.74 years of education compared to 14.49 years of schooling among greenhouse producers. Some open field farmers had 0

Table 1. Descriptive analysis of continuous characteristics.

Variable	Sample	Open field (n=306)			Greenhouse (n=78)			t-value	Sig
	N=384	Mean	Min	Max	Mean	Min	Max		
Age (Years)	37.03	36.36	25	75	39.64	25	68	-2.76	.006***
Credit (Ksh)	29930	9998	0	300000	108121	0	500000	-8.40	.000***
Household size (No)	5 .14	5.16	1	10	5.08	2	10	0.34	.734
Experience (Years)	9.07	10.55	1.5	25	3.26	2	5	21.2	.000***
Education (Years)	9.90	8.74	0	16	14.49	10	18	-23.3	.000***
Land size (ha)	0.709	0.649	0.09	2	0.95	0.18	1.80	-4.99	.000***
Farm size (ha)	2.30	2.294	1.60	8.40	2.32	1.60	4.9	-0.29	.766
Fertilizer (Kg/ha)	208.8	236.5	18.5	1200	99.68	20	350	10.0	.000***
Seeds (g/ha)	46.87	54.21	2.50	300	18.10	2.78	138.9	9.34	.000***
Pesticides (L/ha)	8.0	8.34	1	48	6.69	1.33	29.2	1.99	.048**
Labour (Mds/ha)	303.7	349.7	42.2	2175	122.8	35	545	11.4	.000***
Productivity(Kg/ha)	8225	7046.	556	23480	12851	3055	21600	-7.94	.000***
Market distance (Km)	9.72	11.11	3	28	4.25	2	8.5	19.0	.000***

***Significance at 1%; ** Significance at 5%.
Source: Field survey data (2019).

years of education implying that they had no formal education. Years spent in school by farmers in either systems were different at 1%.This exhibited that greenhouse farmers were significantly more educated thus had enhanced skills and ability to better utilize market information and understand modern technologies.

On average, the respondents were located 9.72 km from the markets. Open field and greenhouse producers were situated 11.11 and 4.25 km from the nearest markets, respectively. Differences in market distances between open field and greenhouse farmers were statistically different implying that open field farmers were located farther away from the markets compared to greenhouse farmers. Further, this shows that greenhouse farmers had adequate access to market information and market benefits on provision of key inputs such as improved seeds and fertilizers. Regarding farm sizes, respondents had an average of 2.30 ha while land size under tomatoes averaged at 0.7096 ha. This shows that the farms were highly fragmented and that tomato production faced competition from other farm enterprises. While farm sizes between open field and greenhouse tomato producers were insignificant, the differences in size of land planted with tomatoes between the two production systems were significant at 1% level. This implies that on average, area under tomato cultivation for greenhouse farmers was considerably large than that of the open field farmers thus expected to give higher outputs.

As regards fertilizer application, di-ammonium phosphate (DAP) and nitrogen, phosphorus, potassium (NPK) fertilizers were the most common during land preparation and planting. Urea and calcium ammonium nitrate (CAN) were frequently used during top dressing. Respondents used a mean of 208.8 kilograms of fertilizer per hectare. From the results, open field farmers used significantly more fertilizer quantity per hectare than the greenhouse producers. The recommended levels of fertilizers in tomato production are approximately 1,186 kilograms per hectare (Tabe and Molua, 2017). Compared with the amounts applied in Kirinyaga, farmers used less than the recommended fertilizer amount.

Seed quantity averaged 46.87 g per hectare and significantly varied between open field and greenhouse tomato farmers in the study area. The most common pesticides Were ridomil and Milraz (fungicides), Karate and Bestox (insecticides) and Oxy gold (herbicide). On average farmers used 8.0 litres of pesticides during the season with 8.34 L per hectare for open field and 6.69 L per hectare for greenhouse farmers. Pesticide application between the two systems differed significantly at 5% level with open field farmers using more per unit of land compared to the greenhouse farmers. This was possibly due to high pest and disease infestation in the open field system.

During the tomato growing season under review, a mean of 303.7 man days per hectare were employed in tomato production. Open field system substantially required more labor in tomato production per hectare (349.76 Mds/ha) compared to greenhouse systems (122.8 Mds/ha). This was attributed to that tomato production under the greenhouse system is highly automated with a drip irrigation which enables distribution of liquid fertilizers and irrigation water thus drastically reducing labour requirements. The mean yield for the sample was 8225 kg/ha (8.225 tons/ha) which was below

Table 2. Descriptive statistics on categorical variables.

Variables	Sample (N=384)		Open field (n=306)		Greenhouse (n=78)		Chi-square test	Sig
	No.	%	No.	%	No.	%		
Gender								
Male	291	75.8	231	75.5	60	76.9	0.070	0.792
Female	93	24.2	75	24.5	18	23.1		
Group membership								
No	240	62.5	188	61.4	52	66.7	0.725	0.394
Yes	144	37.5	118	38.6	26	33.3		
Land tenure								
Without title	196	51.1	159	51.9	37	47.4	0.509	0.475
With title	188	48.9	147	48.1	41	52.6		
Type of seed								
Uncertified	167	43.5	167	54.9	0	0	76.94	0.000***
Certified	215	56.5	137	44.8	78	100		
Extension								
No	300	78.1	245	80	55	70.5	3.319	0.068**
Yes	84	21.9	61	20	23	29.5		
Market information								
No	26	6.8	22	7.2	4	5.1	0.418	0.518
Yes	358	93.2	284	92.8	74	94.9		
Total	384	100	306	100	78	100		

***Significance at 1%; ** Significance at 5%.
Source: Field survey data (2019).

a potential of 30.7 tons per hectare (Wachira et al., 2014). The average productivity was significantly different between production systems with 7046.57 kg per hectare (7.05tons/ha) for open field and 12850.47 kg per hectare (12.85tons/ha) for greenhouse. The greenhouse system was more productive than the open field system in tomato production among farmers in the sample. However, this productivity remained low compared to 23 tons per hectare for open field and 161 tons per hectares for greenhouse system (Van der Spijk, 2018).

Categorical farm and farmer characteristics

Table 2 gives a comparison of categorical factors of respondents in the sample. From Table 2, of the sampled household heads, 306 (79.68%) grew tomatoes under the open field system while 78 (20.32%) adopted the greenhouse production system. The low adoption could be attributed to limited knowledge on emerging innovations in tomato production and high initial cost of investments required to establish greenhouse structures.

Majority (75.78%) of the sampled households were male headed, with only 24.22% being female headed. However, the results show that there was no statistically significant relationship between gender and the type of system used. Concerning farmers groups, only 37.5% of the respondents had group membership but the connection between group membership and production systems was not statistically different. The results show that 51.04% of the respondents owned land with title deeds while 48.96% operated farms that were either leased, communally owned or had permission to use from the land owners. Linkages between land tenure and the two production systems did not differ significantly. Further, relations between seed type used by the respondents in either production systems were statistically different as shown by the chi-square value. All the sampled greenhouse producers used certified seeds with a sizeable proportion of the open field farmers using uncertified seeds. This was motivated by allocation of credits in tomato production which enabled farmers to timely procure of improved seeds for production.

Access to extension services was limited in the study with only 21.87% of the respondents having contact with extension agents. Similar results were obtained within systems, with only 20% of the open field farmers and 29.5% of the greenhouse farmers having access to extension and training. The relation between access to extension and type of tomato production systems differed at 5% level. This implies that farmers most of the green-house farmers were adequately trained better agricultural

Table 3. Maximum likelihood estimates of the stochastic production function.

Variable	Parameter	Coefficient	Std. error	z	P >|z|
Constant	β_0	1.7133	0.5244	3.27	0.001***
Land size (Hectares)	β_1	0.5917	0.0535	11.07	0.000***
Fertilizer (Kilograms)	β_2	0.4761	0.0748	6.36	0.000***
Seed quantity (Grams)	β_3	-0.1089	0.0508	-2.14	0.032**
Chemicals (Litres)	β_4	0.0617	0.0579	1.07	0.287
Labour (Man days)	β_5	-0.0336	0.0583	-0.58	0.564
Log likelihood		-447.5662			0.000***
Wald chi2(5)		472.13			0.000***
Lambda		10.7508	0.0923	116.42	0.000***
Likelihood ratio (5, 5%)		15.1389			0.000***
Sigma squared (σ^2)		2.097			0.000***
Gamma (γ)		0.6876			0.000***

***Significance at 1%; ** Significance at 5%.
Source: Field survey data (2019).

techniques and technologies that have potential to increase yields. Majority (93.23%) of the respondents were privy to trends in both input and output markets with 92.8% open field and 94.9% greenhouse farmers having access to market information. Relations between access to market information between farmers in either open field or greenhouse system did not differ significantly.

Parametric estimates of frontier production function

The maximum likelihood estimation procedures were used to estimate the stochastic Cobb-Douglas production frontier function using STATA software and the results is given in Table 3.

The value of gamma parameter (γ) shows that 68.76% of the deviations in tomato production resulted from technical inefficiencies. The results yielded a sigma squared (σ^2) value of 2.097 that was significant at 1% level. This denotes a perfect goodness of fit with the Cobb Douglas stochastic frontier model. The value (15.1389) of the likelihood ratio (LR) test was significant at 1% level and greater than the critical value of chi-square (11.070) with 5 degrees of freedom. This shows that the Cobb Douglas functional form was appropriate for data. The results show that area under tomato cultivation (land size), fertilizer quantity applied and seed quantity used were important in determining tomato production in the study area.

Acreage under tomato cultivation (land size) and fertilizer quantity had positive coefficients that were significant at 1% level. Thus increasing acreage under tomato production and fertilizer usage by 1% would increase tomato output by 0.5917 and 0.4761%, respectively. In addition, seed quantity had a negative and significant coefficient at 5% level showing that a 1% increase in seed quantity would reduce tomato output by 0.1089%. This is reasonably due to use of local uncertified seeds by a sizeable proportion of respondents that contain high levels of impurities which reduce the germination potential. This reduces the plant population per unit of land leading to low yields. These result concurred with Wabomba (2015). Output was highly responsive to tomato acreage, followed by fertilizer and seed quantity.

Comparison of technical efficiency in open field and greenhouse production systems

Table 4 shows the distribution of technical efficiency scores of the sample and the comparison of the efficiency between open field and greenhouse tomato production systems.

The mean technical efficiency for the sample was 39.55. This shows that there exists an opportunity to improve technical efficiency by more than 60% if all restrictions that make smallholder tomato farmers in Kirinyaga County inefficient are improved. The results coincided with the findings of Zalkuw et al. (2014). The mean technical efficiency for open field farmers was 31.48% and 71.22% for the greenhouse farmers. This implies that greenhouse farmers had a higher technical efficiency value compared to that of open field farmers. This result negated the findings of Najjuma (2016) who estimated mean technical efficiency of 40.43 and 33.71% for open field and greenhouse farmers, respectively. In addition, technical efficiency ranged from 3.63% to 94.62%. The wide range indicates that most of the smallholder farmers utilized available resources inefficiently.

Majority (80.4%) of the open field farmers had efficiency levels below 50% with only 12.8% greenhouse

Table 4. Frequency distribution of technical efficiency.

Description	Efficiency range	Sample		Open field		Greenhouse	
		No.	%	No.	%	No.	%
Low	0 < to < 0.25	159	41.4	158	51.6	1	1.3
Moderately low	0.25 < to < 0.50	97	25.3	88	28.8	9	11.5
Moderately high	0.50 < to < 0.75	68	17.7	40	13.1	28	35.9
High	0.75 < to ≤ 1.00	60	15.6	20	6.5	40	51.3
Mean		0.3955		0.3148		0.7122	
Minimum		0.0363		0.0362		0.9462	
Maximum		0.9462		0.1536		0.9361	
Standard deviation		0.2667		0.2220		0.1763	

Source: Field survey data (2019).

Table 5. One way ANOVA comparison of technical efficiency.

Technical efficiency	Sum of squares	df	Mean squares	F	Sig
Between groups	9.816	1	9.816		
Within groups	17.433	382	0.046	215.098	0.000***
Total	27.249	383			

***Significance at 1%.
Source: Field survey data (2019).

farmers below this level. The results also noted that 66.7% of the smallholder tomato farmers had efficiency scores below 0.5. This implies that by increasing technical efficiency, 66.7% of the sampled farmers could increase tomato output by more than 50% with majority being open field farmers. The results agreed with the studies of Khan and Shoukat (2013) in northern Pakistan, Ayerh (2015) in Ashanti region of Ghana. Within the production systems, only 19.6%of open field and 87.2% greenhouse farmers, attained efficiency levels of 50% and above. In addition, 33.3% of the farmers attained efficiency levels of 50% and above.

The variations observed in technical efficiency between farmers in open field and greenhouse tomato production systems were statistically different at 1% level. This confirms that among smallholder tomato farmers in Kirinyaga County, greenhouse system of production was significantly more technically efficient than the open field system as shown in Table 5. The plausible explanation is that farmers who use greenhouses adopted certified seeds and were significantly more educated thus understood the role of modern technologies in production.

Socio-economic and institutional factors affecting technical efficiency

The effect of selected factors on technical efficiency was ascertained using censored Tobit regression model as specified in Equation six (6). Table 6 shows the results of Tobit multiple regression analysis. The existence of inefficiency was determined using the log likelihood which gave a value of 88.22 that was significant at 1% level. The Tobit regression denoted a likelihood ratio (LR) of 250.27. The critical value of chi-square (27.857) at 5% level of significant with 17 degrees of freedom was less than the LR. This denotes that the Tobit regression model was appropriate in determining factors that affect technical efficiency in the study area.

Households size had a significant coefficient at 1% level and positively influenced technical efficiency. This implies that as the households size expands, technical efficiency among smallholder tomato farmers in the study area increases. This implies that farmers with large households are more technically efficient compared to farmers whose households are small. The plausible explanation is that big households strive to meet their subsistence thus endeavor to achieve higher outputs.

Further, since tomato production is labour intensive, large household afford labor endowments necessary to execute farm decisions. The results coincided with the studies of Ayerh (2015) and Ibitoye et al. (2015). On the contrary, the results negated the findings of Folorunso and Adenuga (2013).They argued that households provide family labor which is associated with inefficiency, thus its increase at farm level reduces technical efficiency.

Type of production system, presented as a dummy of

Table 6. Tobit regression results on factors affecting technical efficiency.

Variable	Coefficient	Std. error	t	p>/t/
Age (Years)	-0.000835	0.001360	-0.61	0.540
Gender (0= Male, 1= Female)	-0.023283	0.023651	-0.98	0.326
Household size (Number)	0.019196	0.005652	3.40	0.001***
Group membership (0=No, 1=Yes)	0.028206	0.021185	1.33	0.184
Experience (Years)	0.000163	0.025128	0.06	0.949
Education (Years)	-0.003479	0.003729	-0.93	0.351
Type of system (0=open field, 1= Greenhouse)	0.446175	0.047588	9.38	0.000***
Land tenure (0=without title, 1=with title)	0.006994	0.091979	0.33	0.740
Seed type (0=uncertified, 1= certified)	0.043299	0.022004	1.97	0.050**
Off farm income (Kenyan shilling)	1.45e-06	1.01e-06	1.44	0.150
Farm income (Kenyan shilling)	7.92e-08	1.43e-07	0.55	0.580
Land size (Hectares)	-0.15262	0.022399	-6.81	0.000***
Fertilizer used (Kilograms)	0.000754	0.000241	3.14	0.002***
Extension (0=No, 1=Yes)	0.041649	0.025128	1.66	0.098
Market distance (Kilometres)	-0.00291	0.002152	-1.35	0.178
Market information access (0=No,1=Yes)	0.078295	0.042304	1.85	0.065
Credit value (Kenyan shilling)	-2.98e-07	2.03e-07	-1.47	0.143
Constant	0.205409	0.091979	2.23	0.026

Log Likelihood = 88.22***; Likelihood Ratio (LR) = 250.27***.***Significance at 1%; ** Significance at 5%.
Source: Field survey data (2019).

open field and greenhouse systems had a positive and significant coefficient. This concurred with prior anticipations and inferred that by farmers embracing greenhouse systems, technical efficiency increased. The likely explanation is that greenhouse system enables prolonged cultivation hence increased tomato production. Further, adverse climatic conditions are largely controlled in the greenhouses which lead to increased yields. Additionally, greenhouse farmers used certified seed in production with majority of the farmers in this system engaging in mono-cropping, thus adequate time and resources are allocated with reduced nutrient competition. The results concurred with Wachira et al. (2014). Further, the results drew a discrepancy with the findings of Najjuma (2016) who reported that due to under exploitation of technologies, open field system was more efficient in tomato production compared to the greenhouses in Kiambu County.

Type of seed used in production was significant at 5% and positively related to technical efficiency. This suggests that by using certified seeds, technical efficiency among smallholder tomato farmers increased. This was facilitated by a sizeable proportion of the respondent apportioned credits in tomato production which enabled them to timely procure improved seeds. In addition, majority of the farmers had adequate information regarding the role of markets in the provisions of affordable inputs. These aspects allowed timely uptake of techniques that increase technical efficiency and efforts in research to generate improved planting materials. The results agreed with the findings of Tasila et al. (2019) and Mukhtar et al. (2018).However, the findings differed with those of Abdul and Isgin (2016) who found an inverse relation between improved seeds and technical efficiency.

The area under tomato cultivation portrayed a significant coefficient at 1% level and was inversely related to technical efficiency. This implies that farmers with small land sizes were more technically efficient than farmers with large plots of land. The reasonable justification is that, farmers with small land sizes give more attention to their farms since they depend on farming for occupation. This prompts them to be more committed in farming and ensure prudent resource combination thus reducing inefficiencies. This result coincided with a study by Dessale (2019) negated the findings of a study by Ibitoye et al. (2015) who found a positive relation between land under cultivation and technical efficiency. In addition, fertilizer quantity had a significant coefficient at 1% level and positively influenced technical efficiency. This positive effect symbolizes that, increased fertilizer application in the study area increased technical efficiency among smallholder tomato producers. This is possibly due the fact that the nutritional composition of fertilizers upgraded soil fertility an element that is of utmost importance in tomato production. The results agreed with the findings of Shettima et al. (2015).

Conclusion

A number of studies on technical efficiency have been conducted in developing countries. However, studies on technical efficiency of tomato farmers in Kirinyaga County have been limited. Therefore, this study sought to estimate technical efficiency and identify characteristics that affect technical efficiency among smallholder tomato farmers in Kirinyaga County of Kenya. The results shows that farmers inefficient with a mean technical efficiency of 39.55% with greenhouse farmers more technically efficient compared to open field farmers. The distribution of efficiency scores ranged from 0.0363to 0.9462 with majority of the households below 50% and 15.6% above 75% efficiency level. This shows a wide range in the technical efficiency scores and a chance to increase technical efficiency by more than 60% if all restrictions that make smallholder tomato farmers inefficient are improved.

Recommendations

Given that the findings of the study showed that technical inefficiencies existed in tomato production among smallholder farmers in Kirinyaga County, the study developed the following recommendations to guide farmers and policy makers in increasing tomato production and productivity at farm level:

i) The farmers should embrace use of certified seeds which are disease resistant and possess high yielding potential. This will reduce cost on pesticides application and ensure high yields. In addition, farmers should apply fertilizers at recommended levels of 1,186 kilograms per hectare since fertilizer application has been found to increase tomato output. This will ensure high yields and better net returns.

ii) The County Government of Kirinyaga should ensure enhanced accessibility of extension services to educate smallholder farmers on emerging innovations and technologies in tomato production. The Kenyan Government should develop policy interventions geared towards subsidizing the costs of establishing greenhouse structures and stabilizing the factor prices of key inputs such as fertilizers and certified seeds. This is because they were found to be important components towards increased technical efficiency levels in tomato production.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Abdul MM, Isgin T (2016). Technical Efficiency of Cassava Production in the Savannah Zone of Northern Ghana: Stochastic Frontier Analysis. Journal of Biology, Agriculture and Healthcare 6(20):62-72.

Aigner DJ, Lovell CK, Schmidt P (1977). Formulation and Estimation of Stochastic Frontier Production Function Models. Journal of Econometrics 6(1):21-37.

Ajibefun IA (2008). An evaluation of parametric and non-parametric methods of technical efficiency measurement: application to small scale food crop production in Nigeria. Journal of Agriculture and Social Sciences 4(3):95-100.

Ayerh F (2015). Technical Efficiency of Tomato Farmers in the Ashanti Region of Ghana (Doctoral dissertation, University of Ghana).

Battese GE, Coelli TJ (1995). A model for technical inefficiency effects in a stochastic frontier production function for panel data. Empirical Economics 20(2):325-332.

Bhattarai D, Singh SB, Baral D, Sah RB, Budhathoki SS, Pokharel PK (2016). Work-related injuries among farmers: a cross-sectional study from rural Nepal. Journal of Occupational Medicine and Toxicology 11(1):48.

Chepng'etich E, Nyamwaro SO, Bett EK, Kizito K (2015). Factors that influence technical efficiency of sorghum production: A case of small holder sorghum producers in Lower Eastern Kenya. Advances in Agriculture 2015:1-11.

Dessale M (2019). Analysis of technical efficiency of small holder wheat-growing farmers of Jamma district, Ethiopia. Agriculture and Food Security 8(1):1.

Folorunso ST, Adenuga KM (2013). An analysis of technical efficiency of ginger crop production in Jaba Local Government Area, Kaduna State, Nigeria. Advances in Applied Science Research 4(5):85-90.

Geoffrey SK, Hillary NK, Kibe MA, Mariam M, Mary MC (2014). Challenges and strategies to improve tomato competitiveness along the tomato value chain in Kenya. International Journal of Business and Management 9(9):205.

Government of Kenya (GoK) (2018).Kirinyaga County Government. County Integrated Development Plan 2018-2022.

Ibitoye SJ, Shaibu UM, Omole B (2015). Analysis of resource use efficiency in tomato (SolanumLycopersicum) production in Kogi state, Nigeria. Asian Journal of Agricultural Extension, Economics and Sociology pp. 220-229.

Kassa MD, Demissie WM (2019). Smallholders' technical efficiency of teff production in Ethiopia. African Journal of Agricultural Research 14(33):1641-1648.

Katungwe F, Elepu G, Dzanja J (2017). Technical Efficiency of Smallholder Tea Production in South-Eastern Malawi: A Stochastic Frontier Approach. Journal of Agricultural Sciences–Sri Lanka 12:3.

Khan REA, Shoukat G (2013). Technical efficiency of tomato production: a case study of district Peshawar (Pakistan). World Applied Sciences Journal 28(10):1389-1392.

Kumar N, Bharat D, Amaresh N, Shivakumar H, Shivakumar R, ArshadP, Subramanian R, Easdown W, Bindumadhava H, Nair RM (2018). Science-based horticultural interventions for improving vegetable productivity in the state of Karnataka, India. Cogent Food and Agriculture 4(1):1461731.

Mitra S, Yunus M (2018). Determinants of tomato farmers' efficiency in Mymensingh district of Bangladesh: Data Envelopment Analysis approach. Journal of the Bangladesh Agricultural University 16(1):93-97.

Ministry of Agriculture (MoA) (2011). Ministry of Agriculture, Livestock and Fisheries Strategic Plan 2013-2017. Kenya Vision 2030.Towards an innovative, commercially Oriented and Modern Agriculture. Nairobi, Kenya

Mukhtar U, Mohamed Z, Shamsuddin MN, Sharifuddin J, Iliyasu A (2018).Application of data envelopment analysis for technical efficiency of smallholder pearl millet farmers in Kano state, Nigeria. Bulgarian Journal of Agricultural Science 24(2):213-222.

Mwangi MW, Kimenju JW, Narla RD, Kariuki GM, Muiru MW (2015). Tomato management practices and diseases occurrence in Mwea West Sub County. Journal of Natural Sciences Research 5(20):119-124.

Narcisse M (2017). Socio-economic factors affecting technical efficiency of small holder's maize production in Rwanda (Doctoral dissertation).

NajjumaE (2016). Techinical Efficiency of Tomato Production : A Comparative Study between Small Scale Green House and Open field tomato farmers in Kiambu. Journal of Agriculture Policy 1, 2(1):16-36.

Najjuma E, Mbeche R, Kavoi MM (2016). Assessment of technical

efficiency of open field tomato production in Kiambu County, Kenya (stochastic frontier approach). Journal of Agriculture, Science and Technology 17(2):21-39.
Ndirangu SN, Mbogoh SG, Mbatia OLE (2018). Evaluation of the Elasticity of Farm Output among Smallholder Farmers in Selected Agro-Ecological Zones of Embu County, Kenya. Asian Journal of Agricultural Extension, Economics and Sociology pp. 1-10.
Nyamwamu RO (2016). Implications of human-wildlife conflict on food security among small holder agropastoralists: a case of smallholder maize (*Zea mays*) farmers in Laikipia County, Kenya. World Journal of Agricultural Research 4(2):43-48.
Okonya JS, Kroschel J (2015). A cross-sectional study of pesticide use and knowledge of smallholder potato farmers in Uganda.BioMed Research International 2015.
Ochilo WN, Nyamasyo GN, Kilalo D, Otieno W, Otipa M, ChegeF, Teresia K, Lingeera EK. (2019). Characteristics and production constraints of smallholder tomato production in Kenya. Scientific African 2:e00014.
Parman BJ, Featherstone AM (2019). A comparison of parametric and nonparametric estimation methods for cost frontiers and economic measures. Journal of Applied Economics 22(1):59-84.
Saavedra TM, Figueroa GA, Cauih JGD (2017). Origin and evolution of tomato production Lycopersiconesculentum in México.Ciência Rural 47:3.
Shettima BG, Amaza PS,Iheanacho AC (2015). Analysis of Technical Efficiency of Irrigated Vegetable Production in Borno State, Nigeria. Journal of Agricultural Economics, Environment and Social Sciences 1(1):88-97.
Simwaka K, Ferrer S, Harris G (2013). Analysis of factors affecting technical efficiency of smallholder farmers: Comparing time-varying and time-invariant inefficiency models. African Journal of Agricultural Research 8(29):3983-3993.
Singh MC, Singh JP, Pandey SK, Mahay D, Srivastava V (2017). Factors Affecting the Performance of Greenhouse Cucumber Cultivation: A Review. International Journal of Current Microbiology and Applied Sciences 6(10):2304-2323.
Tabe OMP, Molua EL (2017). Technical efficiency of smallholder tomato production in semi-urban farms in Cameroon: A stochastic frontier production approach. Journal of Management and Sustainability 7:27.
Tasila KD, Mabe FN, Oteng FR (2019). Profitability and Profit Efficiency of Certified Groundnut Seed and Conventional Groundnut Production in Northern Ghana: A Comparative Analysis. Cogent Economics and Finance P. 1631525.
Tirra AN, Oluoch KW, Nyanganga H, Mwang'ombe AW (2019).Factors influencing the level of commercialization among smallholder cassava farmers in Taita-Taveta and Kilifi Counties, Kenya. African Journal of Agricultural Research 14(32):1584-1592.
Wabomba OW (2015). Determinants of technical efficiency of soybean production among farmers in Bungoma County, Kenya (Doctoral dissertation, Moi University).
Wachira JM, Mshenga PM, Saidi M (2014). Comparison of the profitability of small-scale greenhouse and open-field tomato production systems in Nakuru-North District, Kenya. Asian Journal of Agricultural Sciences 6(2):54-61.
Wahid U, Ali S,Hadi NA (2017). On the Estimation of Technical Efficiency of Tomato Growers in Malakand, Pakistan.Sarhad Journal of Agriculture 33(3):357-365.
Wambua DM, Ndirangu SN, Njeru LK, Gichimu BM (2019). Effects of recommended improved crop technologies and socio-economic factors on coffee profitability among smallholder farmers in Embu County, Kenya. African Journal of Agricultural Research 14(34):1957-1966.
Wanjiku GG (2015). Post-harvest Fungi Diversity and Level of Aflatoxin Contamination in Stored Maize: Cases of Kitui, Nakuru and Trans-nzoia Counties in Kenya (Doctoral dissertation, Kenyatta University).
Van der Spijk CA (2018). Greenhouse technology adoption among small and medium-scale tomato farmers in Kenya.
Yabs JK, Awuor EO (2016). Market Orientation and Performance of Fruit Exporting Firms in Kenya: A Theoretical Perspective. European Journal of Business and Management 8(9)23-33.
Zalkuw J, Singh R, Pardhi R, Gangwar A (2014). Analysis of technical efficiency of tomato production in Adamawa State, Nigeria. International Journal of Agriculture, Environment and Biotechnology 7(3):645.

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Effects of land use on soil physicochemical properties at Barkachha, Mirzapur District, Varanasi, India

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Types of land use practice significantly affect the soil physico-chemical properties. Four different land use types were selected (natural forest, bamboo plantation, degraded forest and agricultural land) to analyze the effect of land uses change on soil chemical and physical properties. Among all land use pattern, the highest water holding capacity (40.06±0.74%), porosity (0.539±0.011%), soil macro-aggregates (64.16±2.64%), soil organic carbon (0.84±0.054%) and soil total nitrogen (0.123±0.013%) were found to be under natural forest, closely followed in decreasing order by bamboo plantation, degraded forest and agricultural land. Unlikely, bamboo plantation was higher in moisture content (2.78±0.23%), whereas agricultural land was lower in moisture content (2.14±0.5%), though no significant differences were observed among land use types. Soil organic carbon was significantly affected by different land use practices. In contrast to this, agricultural land was higher in bulk density (1.37±0.0193 g/cm^3) whereas natural forest was lower in bulk density (1.220±0.0288 g/cm^3). Bulk density, soil organic carbon, soil total nitrogen, water holding capacity and porosity were significantly affected by land use changes. Furthermore, the correlation of analysis showed that soil organic carbon, soil total nitrogen, moisture content, porosity, water holding capacity, soil macro aggregates were positively correlated to each other and negatively correlated with bulk density, meso and micro soil aggregates at p<0.05. The results of this study will help to develop future plan about land use and soil management regarding soil carbon dynamics and climate change mitigation.

Key words: Land use type, land use change, soil physico-chemical properties.

INTRODUCTION

Land use change is one of the major drivers of global environmental change associated mainly with climate change, loss of biodiversity, reduction of soil fertility and changes in ecosystem services (Tilman et al., 2001;

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Ashagrie et al., 2007). It has a deep effect on soil organic carbon (SOC) storage, since it affects the amount and quality of litter input, litter decomposition rate, and stabilization of SOC. About 1500 Gt carbon (in 1 m soil depth) is reportedly present in soil organic matter, which is the largest of all the active terrestrial carbon pools (Eswaran et al., 2000). Among these, about 136±55 Gt carbon was estimated to be lost from soil organic matter stock due to land use change, thus can subsequently alter soil organic matter dynamics. Therefore, land-use change affects soil organic carbon accumulation and storage in soils, which in turns greatly influences the composition and quality of organic matter (Six et al., 2000; John et al., 2005; Helfrich et al., 2006). Land use change not only affects soil organic carbon but it also affects other nutrient contents of soil viz. Nitrogen, phosphorus etc. Restoration or reclamation of these degraded forests is of major concern and possess great challenge.

Soil organic matter has long been recognized as a determinant constitute of soil physical, chemical, and biological quality in general soil quality. In other words, soil organic matter is critically linked to soil physical and chemical properties (Li et al., 2013). Soil quality is the capacity of a soil to function within ecosystem boundaries to sustain biological productivity, maintain environmental quality, and promote plant and animal health (Doran and Parkin, 1994). Thus, the capacity of a soil to function is often described as soil quality used to assess status of land or soil under various management systems (Ayobi et al., 2011). Soil quality indicator is a measurable soil property that affects the capacity of a soil to perform a specified function (Karlen et al., 1997). Soil properties that are responsive to the land use change are considered as suitable soil quality indicators (Carter et al., 1993). Among others various soil physico-chemical properties are considered as index of soil quality. Monitoring and mediating the negative consequences of land use change while sustaining the production of essential resources has therefore become a major challenge.

Nowadays, the practice of land use conversion, for instance from natural ecosystems to cultivated ecosystems is very common throughout the world (Vagen et al., 2006; Khormali and Nabiollahy, 2009). Maintaining soil quality (which includes various soil physical and chemical properties) is of major importance for any soil management system. Carbon sequestration has now been considered as one of the most effective mechanisms for mitigating the loss of soil carbon and related properties, by slowing or reversing the trend of increasing concentration of carbon dioxide in the atmosphere (Asante et al., 2011). Carbon sequestration in soil that is, accumulation of carbon in soil, refers to taking carbon dioxide from the atmosphere through plants and storing the carbon in the soil in the form of soil organic matter. In other words, carbon sequestration denote transferring atmospheric carbon dioxide into resistant pools with slow turnover and storing it firmly so that it is not released immediately (Lal, 2004).

Forests are one of the major ecosystems responsible for carbon sequestration, which cover about 30% of the land surface, store about 45% of total terrestrial carbon. In undisturbed natural forest ecosystems, the additions and losses of carbon are balanced over time and soil carbon stock reaches a stable equilibrium. Conversion of natural forest to other land use types is the major challenge for maintaining good soil quality. Other factors responsible for degradation of natural forests are excessive harvesting of woody and/or non-woody product of forest, grazing, poor management and other anthropogenic disturbances (ITTO, 2002). These disturbances, in turn, affect soil organic carbon accumulation and storage in soils, which greatly influences the composition and quality of organic matter. Plantation of *Jatropha curcas* (a perennial shrub of Euphorbiaceae family) in degraded land may be a good alternative for the reclamation of these degraded lands as the *J. curcas* is drought resistant and not preferred by the animal (Krishnamurthy et al., 2012). Information on the changes in soil physico-chemical properties due to land use changes are limited in general (Murty et al., 2002; Tripathi and Singh, 2009) and particularly lacking in dry tropics. The major objective of the present study is to analyze the effect of land use change on the potential of soil carbon sequestration in terms of the concentration of soil organic carbon.

MATERIALS AND METHODS

Study area and description of sites

The study was conducted in Baranas Hindu University, Varanasi, India from March to August, 2016. Samples were collected from Barkachha, Mirzapur district. Mirzapur is located at 25.15° N 82.58° E. It has an average elevation of 80 m (265 feet). It is a city in Uttar Pradesh, India, roughly 650 km from both Delhi and Kolkata, almost 89 km from Allahabad and 57 km from Varanasi. It has a population of 233,691 (2011 census). The climate in Mirzapur is warm and temperate. The mean annual temperature and rainfall is 26°C and 975 mm, respectively. The study area was classified into four sites based on their vegetation cover: Natural forest, degraded forest, bamboo plantation and agricultural land.

The forest in the study area is the mixed dry deciduous type dominated by *Acacia catechu* Wild., *Albizia odoratissima* (Benth.), *Acacia nilotica* (L.) *Willd*. *Boswellia serrata* Roxb., *Nyctanthes arbor-tristis* L.*,* with scattered trees of *(Azadirachta indica* Juss) and *Zizyphus glaberrima* Santap. The forest floor was covered with herbaceous vegetation comprising *Ocimum americanum.* L. *Pisum arvense* L., Rhynchosia *minima* (L.) DC., *Cassia sophera*(L.) Roxb., *Acrocephalus indicus* (Burm.f.), *Kuntze.*, *Cynodon dactylon* (L.) and *Oplismenus burmannii* Ritz. The degraded forest site was dominated by *Oryza glaberrima*, *Chrysopogon fulvus* (Spreng.), *Heteropogon contortus* (L.), *Adina cordifolia* Roxb. and scattered trees of *Butea monosperma* (Lamk.). Herbaceous vegetation in the degraded forest was dominated by *Cassia tora* L., *Oldenlandia diffusa. (*Willd.) Roxb., *Sporobolus* spp., *Panicumpsilopodium* Trin.

and *Alysicarpus vaginalis* (L.) DC

Soil sampling techniques

Soil samples were taken from four land use types (NF, DF, BP and AL) from the upper 15 cm depth for studying the impacts of land use change on soil physic-chemical properties. The natural forest was further divided into six sub-sites of 100 m × 100 m. From each sub-site, four soil samples were collected and mixed to represent the single composite sample of each study site. The same procedure was followed for degraded forest, bamboo plantation and agricultural land. The samples were immediately brought to the laboratory and were air dried for further analysis by Allen et al. (1974) and Waksman (1952).

Soil analysis

Soil physico-chemical characteristics (moisture, pH and organic content) were analyzed by standard methods as suggested by Allen et al. (1974) and Waksman (1952). For measuring soil moisture content, 10 g of fresh soil was dried at 105°C in oven to constant weight. Soil moisture content was calculated as:

$$\text{Soil moisture content } (\%) = \frac{\text{Weight of fresh soil} - \text{Weight of dry soil}}{\text{Weight of dry soil}} \times 100$$

For pH: Soil was dissolved in the sterilized distilled water in the ratio 1:5 and then measured the pH by using pH meter. Soil bulk density was determined by removing a known volume of soil using metal tubes and oven drying it at 105°C for 24 h. porosity expresses the relative amount of pore space in the soil. It is not measured directly but is calculated from the bulk density and particle density (Brady and Weil, 1996): it was calculated using the equation:

$$[1 - (Db/Dp)] \times 100$$

where Db = bulk density, Dp = particle density (assumed to be 2.65 Mg m-3 soil). The water holding capacity of the soil was determined using perforated circular brass boxes according to Piper (1966) method. Soil organic C was estimated by the dichromate oxidation and titration method (Kalembasa and Jenkinson, 1973). Total N concentration was measured by the micro kjeldahl method (Jackson, 1973) by using a Gerhardt digester and distillation unit.

Soil aggregates

Soil aggregates were determined by dry method according to Kemper and Chepil (1965). Air dried soil samples (100 g) were placed on a set of seven stacking of sieves and sieved for 3 min on a horizontal shaker (92 rpm), and three dry aggregate size classes separated were,1000 mm (macro-aggregate), 212-500 mm (meso-aggregate) and 53-150 (micro-aggregate).

RESULTS

Physico-chemical properties of the soil

The major physical properties of soils of different sites investigated are presented in Table 1 (pH, moisture content, aggregates, porosity, water holding capacity (WHC) and bulk density (BD)). As shown in Figure 1a, moisture content was highest in bamboo plantation (2.78%); while the lowest value was found in agricultural land (2.14%). However, no significant differences (at $p<0.05$) in moisture content were found among the land use types. The content of moisture in natural forest and degraded forest were 2.32 and 2.25%, respectively. Bulk density was found to vary significantly across the land use types; it was higher in agricultural land (1.37 g/cm^3), followed by degraded forest (1.25 g/cm^3), bamboo plantation (1.223 g/cm^3) and natural forest (1.22 g/cm^3). A significant difference in bulk density was observed between natural forest and other land use types (DF, BP and AL) but no significant different were observed among DF, BP and Al (Figure 1b). Soil porosity followed a reverse trend to that of bulk density. Porosity was significantly affected by land use change; it was higher in natural forest (0.539%) than bamboo plantation (0.538%); whereas the lowest was recorded in degraded forest (0.529%) and agricultural land (0.48%). So, significant difference was observed between natural forest and other land use types; whilst no significant difference was observed among degraded forest, bamboo planation and agricultural land (Figure 1c). The water holding capacity of natural forest, degraded forest, bamboo plantation and agricultural land is displayed in Figure 1d. Higher WHC (40.06%) was found in Natural forest and followed in decreasing order by bamboo plantation (38.51%), degraded forest (37.23%) and agricultural land (29.72%). There was a significant difference between agricultural land and other land use types in water holding capacity; whilst there was no significant difference among natural forest, degraded forest and bamboo plantation at $p<0.05$.

Soil aggregates

Soil aggregate is the naturally occurring cluster or group of soil particles and measures formation of organo-mineral complex (union of mineral and organic matter) in the soil. Across different land use types, macro-aggregates constituted (42-64%) of total soil followed by meso-aggregates (25-336%) and micro-aggregates (10-20%) (Table 2). Macro-aggregates were significantly higher in natural forest (64.16%) followed by bamboo plantation (51.65%), degraded forest (46.83%) and agricultural land (42.94%). Meso and micro-aggregates were higher in agricultural land followed by degraded forest, bamboo plantation and natural forest. The ANOVA results revealed that a significant difference in macro and meso-aggregates between natural forest and the other land use types. The micro-aggregates were also significantly affected by land use change; natural forest was significantly different with degraded forest and agricultural land but no significant difference with bamboo plantation.

Chemical properties of the soil

Results of the soil chemical properties (particularly

Table 1. Percentage of distribution of different dry aggregate soil size classes in different land use types and soil organic carbon (SOC) and soil total nitrogen (STN.

Soil aggregates (%)	Land use type				LSD
	NF	DF	BP	AL	
Macro-aggregates	64.16±2.64[a]	46.83±1.00[b]	51.65±5.4[b]	42.94±1.11[b]	8.48
Meso aggregates	25.68±2.48[a]	35.16±0.73[b]	33.10±4.74[b]	36.39±1.69[b]	9.28
Micro-aggregates	10.16±1.38[a]	18.01±0.96[b]	15.25±1.86[ba]	20.66±1.81[b]	4.65
SOC (%)	0.84±0.054[a]	0.448±0.113[b]	0.72±0.074[a]	0.435±0.042[b]	0.21
STN (%)	0.123±0.013[a]	0.027±0.003[b]	0.033±0.0034[b]	0.014±0.0016[b]	0.021

Values are mean ± SE. In each rows, values having different superscript are significantly different from each other ($p < 0.05$).

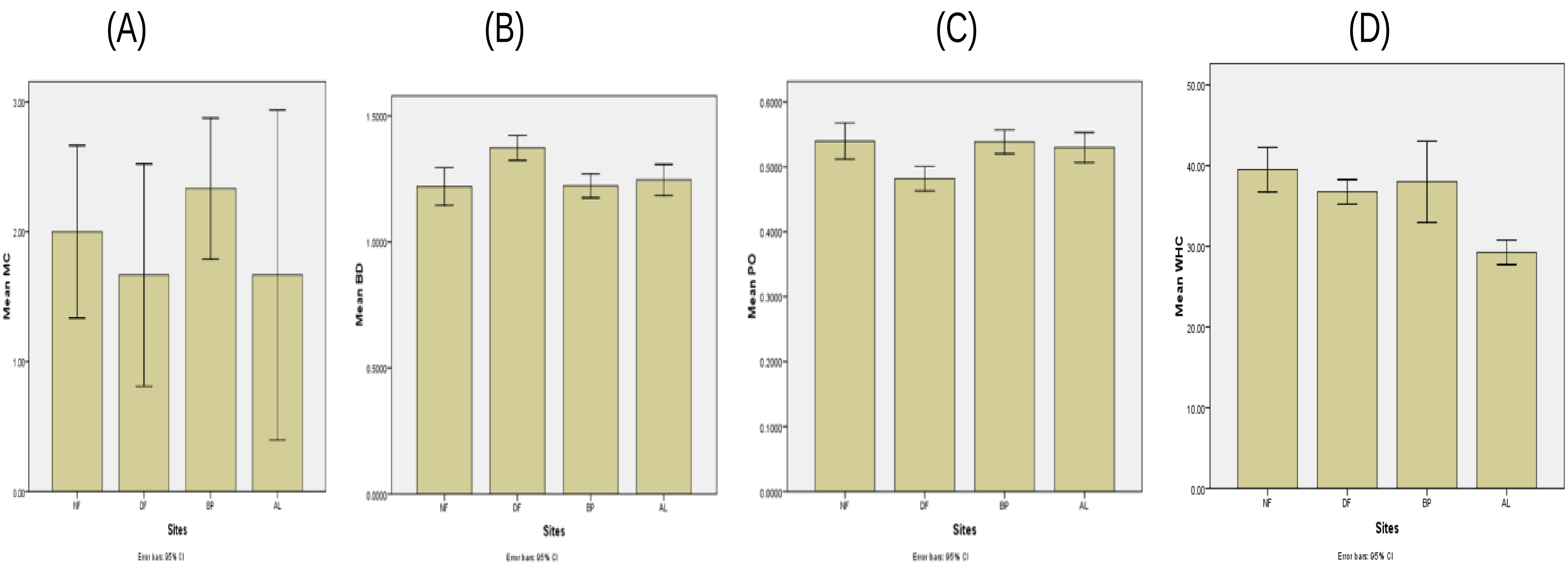

Figure 1. Comparison of MC, BD, porosity and WHC under different land use types: Natural Forest (NF), Degraded Forest (DF), Bamboo Plantation (BP) and agricultural Land (AL).

organic carbon and total nitrogen (tN)) of natural forest, degraded forest, bamboo plantation and agricultural land are presented in Table 1. Soil organic carbon and total nitrogen varied considerably across the land use types. The highest soil organic carbon was obtained from

Table 2. Correlation matrix for physical and chemical characteristics of soils from different land uses.

Soil variable	SBR	SOC	STN	MC	PO	WHC	BD	MA	ME
SOC	0.997**	1							
STN	0.815	0.828	1						
MC	0.583	0.548	0.01	1					
PO	0.758	0.703	0.555	0.615	1				
WHC	0.808	0.76	0.66	0.547	0.991**	1			
BD	-0.776	-0.722	-0.568	-0.624	-1.000**	-0.992**	1		
MA	0.926	0.929	0.970*	0.251	0.703	0.787	-0.717	1	
ME	-0.89	-0.901	-0.989*	-0.154	-0.623	-0.718	0.637	-0.994**	1
MI	-0.951*	-0.946	-0.936	-0.35	-0.778	-0.848	0.79	-0.993**	0.973*

BD, Bulk density; WHC, Water holding capacity; SOC, Soil organic carbon; STN, Total nitrogen; PO, Porosity; MA, Macro aggregates; ME, Meso aggregates; MI, Micro aggregates. Pearson's correlation coefficient, n = 4, * p < 0.05, ** p < 0.0.

natural forest (0.84%) followed by bamboo plantation (0.72%), degraded forest (0.448%) and agricultural land (0.435%). Soil organic carbon in natural forest and bamboo planation were significantly different with agricultural land and degraded forest; whilst there was no significant difference recorded between natural forest and bamboo planation, and degraded forest and agricultural land. Similarly, variation in soil total nitrogen concentration along the various land use types was found to be highest in natural forest (0.123%) followed in decreasing order bamboo plantation (0.033%), degraded forest (0.027%) and agro-ecosystem (0.014%). The analysis of variance showed that there was significant difference between natural forest and other land use types at $p<0.05$ in soil total nitrogen. However, no significant differences were observed among degraded forest, bamboo plantation and agricultural land.

Pearson's correlation coefficients between SOC, STN, moisture content, porosity, WHC, soil aggregates and bulk density is given in Table 2 (Singh and Ghoshal, 2014). SOC, STN and soil macro aggregates were strongly positively correlated to each other, and negatively correlated with bulk density, meso and micro soil aggregates. Additionally, soil organic carbon and total nitrogen were positively correlated with porosity (r=0.703 and 0.555), water holding capacity (r=0.76 and 0.66) and macro soil aggregates (r=0.929 and 0.970) while less/weakly correlated with moisture content (r=0.548 and 0.01). By contrast, soil organic carbon and nitrogen negatively correlated with bulk density (r=-0.722, and -0.568, $p<0.05$, respectively), soil meso (r= -0.901, -0.989, respectively) and micro aggregates (r= -0.946 and -0.936, $p< 0.05$, respectively). Macro soil aggregates was found to be significantly positively correlated with soil total nitrogen (r=0.97, $p<0.05$).

DISCUSSION

As per the finding of this study, the changes of tree species and composition was significantly affected the physical and chemical properties of soil. Natural forest found to be significantly higher in porosity, macro-aggregates, WHC but the least in bulk density, meso-aggregates and micro soil aggregates. In contrast, agricultural land found to be the higher in bulk density as compared to natural forest, bamboo plantation and degraded forest (Figure 1 and Table 1). This result supported by the study conducted by Tripathi et al., (2007) forest and mixed forest ecosystems were possess higher organic matter content compared to savanna and cropland ecosystems. According to Tripathi et al., (2007) soil physical and chemical properties can be significantly improved for the vegetation systems and Chen et al. (2010) land use change may lead to changes in soil physical, biological and chemical properties through their influence on various ecological processes.

Bamboo plantation (2.78%) was higher in moisture content followed by natural forest (2.32%), degraded forest (2.25%) and agricultural land (2.14%) but no statistically significant difference was found among the land use types (Figure 1a). This result was similar with earlier report by Pereira et al. (2013) the moisture of soil in re-forested Araucaria areas was higher as compared to natural forest and crop land. The reason for decreased soil moisture level in the cropland compared to forest ecosystems might be due to the decrease in organic matter and aeration following repeated cultivation, which may promote drying (Singh et al., 2009). Moreover, Singh et al. (2009) reported in cultivated soils, evaporation is a moisture-loss mechanism in the upper soil layer (0-10 cm) and there was about 17% decline in the soil moisture following cultivation. Natural forest was found to be higher in porosity and then in decreasing order bamboo planation, degraded forest and agricultural land (Figure 1c). This is suitable for adequate oxygen diffusion and water infiltration into the soil. This shows a good structural quality, favorable for the successful development of the biological community (Pereira et al., 2013).

Natural forest was significantly higher in WHC (40.06%)

than other three land use types. The WHC of bamboo plantation, degraded forest and agricultural land were (38.51%), (37.23%) and (29.72%). respectively (Figure 1d). This is comparable with other similar study elsewhere. For instance, Singh et al. (2009) reported WHC was greater in forest soils compared to savanna and cropland soils. Soil water is retained in pore spaces and adsorbed onto the surface of mineral and organic matter particles (Li et al., 2007). Cultivation primarily exhausts the labile pool of organic matter, for example, polysaccharides which are hydrophilic, creating a deficiency of adsorbent surfaces within soil and thereby diminishing its WHC (Li et al., 2007). Higher values of porosity in natural forest could be due to more organic matter content and high amount of fine fractions which has a higher surface area (Gupta et al., 2010).

Bulk density was highest in Agricultural land (1.37 g/cm^3) whereas lowest in natural forest (1.22 g/cm^3). The bulk densities of Bamboo plantation and degraded forest were (1.223 g/cm^3) and (1.25 g/cm^3), respectively (Figure 1b). Similarly, Goni et al. (2015) reported higher bulk density was found in wasted land and followed in decreasing order grass land, agricultural land and forest land. Zhang et al. (1988) and Singh et al. (1989) have also reported an increase in soil bulk density due to cultivation. This was probably due to decreased SOC and soil aggregation (Goni et al., 2015), as a result of repeated events of sowing and harvesting. Bot and Benites (2005) also reported that bulk density was lower in soils with high organic matter content.

Land use type displayed significant effect on the aggregate fraction (Table 1). Natural forest had more macro aggregate (64.16%) but lowest in meso (25.68%) and micro aggregates (10.16%). While agricultural land found to be the lowest in macro-aggregates (42.94%) whereas highest in meso (36.39%) and micro aggregates (20.66%). Similarly, in bamboo plantation the macro, meso and micro aggregates were 51.65, 33.10 and 15.25%, respectively; and in degraded forest the macro, meso and micro aggregates were 46.83, 35.16 and 18.01%, respectively. This could be due to the fact that no tillage in natural forest, lower disturbance and higher organic matter input (litters and root exudates) that bind soil aggregates together resulting in improved soil structure formation. By contrast, degraded forest and agricultural land showed lower aggregates due to lower OC content and more micro fraction, respectively attributed to continuous cultivation and rapid oxidation of SOM (Bot and Benites, 2005).

Soil organic C and N is considered to be one of the major attributes of soil fertility and agricultural sustainability (Lal, 2002). As per the finding of this study, the highest organic carbon was found to be under natural forest (0.84%) and the lowest organic carbon was found to be under agricultural land (0.435%). Degraded forest and agricultural land organic carbon were found 0.448 and 0.435%, respectively. Similarly, the highest organic nitrogen was found under natural forest (0.123%) and the lowest organic nitrogen was found under agricultural land (0.014%). Degraded forest and agricultural land organic nitrogen were found 0.027 and 0.033%, respectively (Table 2). This is comparable with other similar studies elsewhere. For instance, Singh and Ghoshal (2011), and Pereira et al. (2013) reported forest had higher in organic carbon and nitrogen than Jatropha plantation/reforested area, degraded forest and lowest in agroe-cosystem; while others reported higher soil organic carbon content in natural forest than tilled crop lands (Gol, 2009). Similarly, Iqbal et al. (2015) also reported the highest soil organic carbon was obtained from agroforestry followed by grass land and fallow land. Highest soil organic carbon concentration in natural forest was might be due to the regular addition of plant litter including above and below ground plant parts, and limited disturbances like grazing, logging, lack of tillage, high plant biodiversity, and root exudates (Goni et al., 2015). Moreover, due to an increased return of residues from high root biomass contributing to the storage and stabilization of SOC in aggregates (Goni et al., 2015; Srivastava and Singh, 1991).

The conversion of natural forest to degraded forest was significantly decreased SOC and tN. The disturbances associated with deforestation might have led to loss of vegetation which in turn have resulted in land degradation, erosion and subsequently the considerable losses of soil organic carbon and nutrients (Tripathi and Singh, 2009; Xiangmin et al., 2014). Changes in land use pattern through changes in type and diversity of plants are reported to exert major influence on the transfer and accumulation of carbon in soil (Tilman et al., 2006). In addition, degradation of natural forest leads to opening of the canopy cover and increases the interference of physical factors such as light intensity, wind velocity and soil moisture content. As the canopy opens, incident light intensity and wind velocity increase, decreasing the moisture content, this, in turn, stimulates organic matter mineralization.

Bamboo plantation was relatively less organic carbon and nitrogen as compare to natural forest, but higher than degraded forest and agricultural land. This increase in soil organic carbon and nitrogen in bamboo plantation was probably due to addition of nutrient rich leaf litter to soil and also due to recycling of these nutrients (Chaudhary et al., 2008; Behera et al., 2010). In contrast to this, the amount of SOC and tN was significantly lower in agricultural land. According to Jenkinson and Rayner (1977), Paul et al. (1997), Tripathi et al. (2007), Saha et al. (2010) studies agricultural practice decreased the level of organic carbon and nitrogen in the soil. Similarly, Poeplau et al., (2010) illustrated that native lands typically stored higher amounts of soil organic carbon than crop lands under similar site conditions because of higher residue inputs and reduced turnover. However, Tripathi et al., 2007) also reported that the SOC and N losses from an agricultural land can be due to its removal of crops. The lowest level of SOC and N from cultivation land may be due to continues tillage practice that accelerates native SOM oxidation by destructing soil aggregates and exposing newer sites to microbial attack which in turn have resulted in loss of SOC (Singh and Ghoshal, 2006).

Conclusion

Soil physico-chemical properties were significantly affected by land use change. Water holding capacity, porosity, soil macro-aggregates, soil organic carbon, and soil total nitrogen were found to be higher in natural forest followed by decreasing order bamboo plantation, degraded forest and agricultural land. In contrast to this, agricultural land was higher in bulk density as compared with other land use types whereas natural forest was lower in bulk density. Soil organic carbon, soil total nitrogen, moisture content, porosity, water holding capacity, soil macro aggregates were positively correlated to each other and negatively correlated with bulk density, meso and micro soil aggregates.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Allen SE, Grimshaw HM, Parkinson JA, Quarmby C (1974). Chemical analysis of ecological materials. in: P.D. Moore, S.B. Chapman (Eds.), Methods in Plant Ecology, Blackwell, Oxford, 1986, pp. 285-344.

Asante P, Armstrong GW, Adamowicz WL (2011). Carbon sequestration and the optimal forest harvest decision: A dynamic programming approach considering biomass and dead organic matter. Journal of Forest Economics 17: 317.

Ashagrie Y, Zech W, Guggenberger G, Mamo T (2007). Soil aggregation, and total and particulate organic matter following conversion of native forests to continuo us cultivation in Ethiopia. Soil and Tillage Research 94:101-108.

Ayobi S, Khormali F, Sahrawat KL, Rodrigues de Lima AC (2011). Assessing Impacts of Land Use Change on Soil Quality Indicators in a Loessial Soil in Golestan Province, Iran. Journal of Agriculture, Science and Technology 13:727-742.

Bot A, Benites J (2005). The importance of soil organic matter: Key to drought-resistant soil and sustained food production. Vialedelle Terme di Caracalla, 00100 Rome, Italy.

Carter MR, Parton WJ, Rowland IC, Schultz JE, Steed GR (1993). Simulation of soil organic carbon and nitrogen changes in cereal and pasture systems of Southern Australia. Australian Journal of Soil Research 31:481-491.

Chaudhary DR, Ghose A, Chikara J, Patolia JS (2008). Soil characteristics and mineral nutrient in wild Jatropha population of India. Communications in Soil Science and Plant Analysis 39:1476-1485.

Chen DD, Zhang SH, Dong SK, Wang XT, Du GZ (2010). Effect of land use on soil nutrients and microbial biomass of an alpine region on the northeastern Tibetan plateau, China. Land Degradation & Development 21:446-452.

Doran JW, Parkin TB (1994). De®ning and Assessing Soil Quality. In: Doran, J.W., Coleman, D.C., Bezdicek, D.F., Stewart, B.A. (Eds.), De®ning soil quality for a sustainable environment. Soil Science Society of America, Madison, pp. 3±21.

Eswaran H, Reich PF, Kimble JM, Beinroth FH, Padamnabhan E, Moncharoen P, Kimble JM (2000). Global carbon stocks. In: Global climate change and pedogenic carbonates, R. Lal, H. Eswaran and B.A. Stewart (eds.), Lewis Publishers, Boca Raton, Florida pp. 15–250.

Gol C (2009). The effects of land use change on soil properties and organic carbon at Dagdami river catchment in Turkey. Journal of Environmental Biology 30:825-830.

Goni R, Sharma N, Iqbal S, Tiwari SC (2015). Soil organic carbon pool under different land uses in Achanakmar Amarkantak Biosphere Reserve of Chhattisgarh, India. Current Science 110:771-773.

Gupta RD, Arora S, Gupta GD, Sumberia NM (2010). Soil physical variability in relation to soil erodibility under different land uses in foothills of Siwaliks in N-W India. Tropical Ecology 51(2):183-197.

Helfrich M, Ludwig B, Buurman P, Flessa H (2006). Effects of land use on the composition of soil organic matter in density and aggregate fractions as revealed by solid-state 13C-NMR spectroscopy. Geoderma 136: 331-341.

Iqbal AM, Hossen SM, Islam NM (2015). Soil organic carbon dynamics for different land uses and soil management practices in Mymensingh Proceedings of 5th International Conference on Environmental Aspects of Bangladesh.

ITTO (2002) 'ITTO guidelines for the restoration, management and rehabilitation of degraded and secondary tropical forest.' ITTO Policy Development Series No 13. (ITTO: Yokohama, Japan).

Jackson ML (1973). Soil Chemical Analysis. Prentice-Hall of India Limited, New Delhi, 574p.

Jenkinson DS, Rayner JII (1977). The turnover of soil organic matter in some of the rothamsted classical experiments. Soil Science 123:298-305.

John B, Yamashita T, Ludwig B, Flessa H (2005). Storage of organic carbon in aggregate and density fractions of silty soils under different types of land use. Geoderma 128:63-79.

Kalembasa SJ, Jenkinson, DS (1973). A comparative study of titrimetric and gravimetric methods for the determination of organic carbon in soil. Journal of Scientific Food Agriculture 24:1085-1090.

Karlen DL, Mausbach MJ, Doran JW, Cline RG, Harris RF, Schuman GE (1997). Soil quality: A concept, definition, and framework for evaluation. Soil Science Society of America Journal 61:410.

Kemper WD, Chepil WS (1965). Size distribution of aggregates. In C.A. Black (ed.) Methods of soil analysis. Part I. Agronomy Monograph No. 9. ASA, SSSA, Madison, WI. pp. 499-509

Khormali F, Nabiollahy K (2009). Degradation of Mollisols in Western Iran as Affected by Land Use Change. Journal of Agricultural Science and Technology 11:363-374.

Krishnamurthy L, Zaman-Allah M, Marimuthu S, Wani SP, Rao AVRK (2012). Root growth in Jatropha and its implications for drought adaptation. Elsevier, Biomass and Bioenergy 39:247-252.

Lal R (2004). Soil carbon sequestration impacts on global climate change and food security. Science 304:16231627.

Lal R (2002). Soil carbon dynamics in crop land and rangeland. Environmental Pollution 116:353–362.

Li W, Yang G, Chen H, Tian J, Zhang Y, Zhu Q, Peng C, Yang J (2013). Soil available nitrogen, dissolved organic carbon and microbial biomass content along altitudinal gradient of the eastern slope of Gongga Mountain. Ecological Society of China. Elsevier 33:266-271.

Murty D, Krischbaum MF, Mcmurtie RE, Mcgilvary H (2002). Does conversion of forest to agricultural land change soil carbon and nitrogen? A review of the literature. Global Change Biology 6:105-123.

Paul EA, Paustian K, Elliott ET, Cole CV (1997). Soil organic matter in temperate agro ecosystems. CRC Press, New York.

Pereira JDM, Baretta D, Bini D, Vasconcellos RF, Cardoso EJBN (2013). Relationships between microbial activity and soil physical and chemical properties in native and reforested Araucaria Angustifolia forests in the state of São paulo, Brazil. Revista Brasileira de Ciencia do Solo 37:572-586.

Piper CS (1966). Soil and plant analysis, Hans Publishers, Bombay.

Poeplau C, Don A, Vesterdal L, Leifeld J (2010). Land use change impact on soil organic carbon stocks: Development of carbon response function from a meta-analysis data set. Geophysical Research Abstracts Vol. 12, EGU2010-10928.

Saha SK, Nair PKR, Nair VD, Kumar BM (2010). Carbon storage in relation to soil size fractions under tropical tree-based land-use systems. Plant Soil 328:433-446.

Singh JS, Raghubanshi AS, Singh RS, Srivastava SC (1989). Microbial biomass acts as a source of plant nutrients in dry tropical forest and savanna. Nature 338:499-500.

Singh JS, Singh DP, Kashyap AK (2009).Comparative account of the microbial biomass-N and N-mineralization of soils under natural forest, grassland and crop field from dry tropical region, India. Plant Soil and Environment 55:223-230.

Singh MK, Ghoshal N (2011). Impact of land use change on soil organic carbon content in dry tropics. Plant Archive 11:903-906.

Singh S, Ghoshal N (2006). Effect of cultivation on major physical and chemical properties of soil. Plant Archives 6:611-613.

Singh MK, Ghoshal N (2014). Variation in soil microbial biomass in the dry tropics: impact of land-use change. CSIRO PUBLISHING; Soil Research 52:299-306.

Six J, Elliott ET, Austian KP (2000). Soil macroaggregate turnover and microaggregate formation: A mechanism for C sequestration under no-tillage agriculture. Soil Biology and Biochemistry 32:2099-2103.

Srivastava SC, Singh JS (1991). Microbial C, N and P in dry tropical forest soils: effects of alternate land uses and nutrient flux. Soil Biology and Biochemistry 23:117-124.

Tilman D, Fargione J, Wolff B, D'Antonio C, Dobson A, Howarth R, Schindler D, Schlesinger WH, Simberloff D, Swackhamer D (2001). Forecasting agriculturally driven global environmental change. Science 292:281284.

Tilman D, Hill J, Lehman C (2006). Carbon-negative biofuels from low-input high diversity grassland biomass. Science 314:1598-1600.

Tripathi N, Singh RS (2009). Influence of different land uses on soil nitrogen transformations after conversion from an Indian dry tropical forest. Catena 77:216-223.

Tripathi N, Singh RS (2009). Influence of different land uses on soil nitrogen transformations after conversion from an Indian dry tropical forest. Catena 77:216-223.

Tripathi SK, Pandey RR, Sharma G, Singh AK (2007). Litter fall, litter decomposition and nutrient dynamics in a subtropical natural oak forest and managed plantation in northeastern India. Elsevier, Science Direct; Forest Ecology and Management: 240:96-104.

Vagen TG, Andrianorofanomezana MAA, Andrianorofanomezana S (2006). Deforestation and cultivation effects on characteristics of Oxisols in the highlands of Madagascar. Geoderma 131:190-200.

Waksman SA (1952). Soil Microbiology, Jhon-Wiley and Sons, New York.

Xiangmin F, Qingli W, Wangming Z, Wei Z, Yawei W, Lijun N, Limin D (2014). Land Use Effects on Soil Organic Carbon, Microbial Biomass and Microbial Activity in Changbai Mountains of Northeast China, Chin. Geogra. Sci.24 :297–306.

Zhang H, Thompson ML, Sandor JA (1988). Compositional differences in organic matter among cultivated and uncultivated Argiudolls and Hapludalfs derived from loess. Soil Science Society of America Journal 52:216-222.

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Full Length Research Paper

Mitigating loan default in Nigeria through joint liability approach: The case of beneficiaries of microfinance bank agricultural loan in Calabar metropolis, Cross River State, Nigeria

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Due to persistent increase in loan default in developing countries especially Nigeria, several lending institutions resort to group lending that is based on joint liability approach to mitigate default. This study analyzes the mitigation of loan default in Nigeria through joint liability approach: The case of beneficiaries of microfinance bank agricultural loan in Calabar metropolis, Cross River State. It specifically analyzes the determinants of loan default among sampled beneficiaries and compares the mean repayment amount between beneficiaries of joint and individual liability. The study used 120 respondents from the selected microfinance institutions in the study area. Data collection was done with the aid of a structured questionnaire. The study used probit model and the Z test to analyze the data. The result revealed that household size and business experience were the key determinants of loan default. There was a significant difference in amounts of loan repaid between joint and individual loan beneficiaries. The study concluded that joint liability is a better approach when it comes to borrowing than the individual liability. The study further shows that the key determinants of loan default were household size and business experience; therefore, beneficiaries are encouraged to reduce their household size as this will reduce default.

Key words: Beneficiaries, default, joint, mitigating, lending, liability, loan, and probit model.

INTRODUCTION

Many studies have shown the importance of credit in promoting economic development and improving household incomes of several countries of the world (Bassey et al., 2016a; Ajah et al., 2014; Enimu et al., 2017). Numerous researchers agreed that the nonexistence of sound - operational credit has been one of the main barriers to the mitigation of worldwide poverty. Intensifying credit availability can assist those who received it to efficiently allocate resources over a period and excellently manage risk, in this way; credit availability can advance profit making chances for the rural poor.

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Though, notwithstanding the observable advantages the rural poor usually find it very challenging to acquire loan. During loan agreement, a lender often needs some form of collateral to protect the loan, in case the borrower is not capable of paying the borrowed funds. Nevertheless, the poor hardly ever gets enough assets to use as collateral. The lender will bear all the loss related with a loan default if there is no collateral from the borrower. To alleviate this problem, hence, proper selection, checking and implementing loan terms is very crucial. In all, these efforts are too expensive for the unacquainted lenders to be sufficiently compensated by interest revenue from very small loans that the poor usually need. In the past few years, micro credit institutions have introduced series of small and non - collateralized loans facilities for the poor. One notable feature of microfinance institution is the joint liability lending and has fascinated significant consideration (Agbaeze and Onwuka, 2014).

In spite of this, poor availability of credit is still predominance as one of the key problems facing small scale business persons in developing countries (Ajah et al., 2017; Bassey, 2014; Bassey et al., 2016a). Other studies such as Ike and Umuedafe (2015) and Ike and Idoge, (2016) lend claims to the inability of formal financial institution to meet the credit requirement of Nigerians.

Certainly, group based microcredit program is one of the most significant innovations in development policy in the last five decades (Mamum et al*.,* 2015). This program permits borrowers who cannot provide security, to come together and be jointly accountable for each other's repayments even though loans are provided to individuals separately. Under such lending conditions, the group takes the responsibility for the individual loans of members and that overcome the problematic issue of unsymmetrical information and consequently the problem of excessively high transaction cost (Agbaeze and Onwuka, 2014; Bassey et al., 2016b).

In Nigeria, credit risk continues to be a peril to microfinance banks sustainability. Many lending institutions in Nigeria especially microfinance banks are confronted by the challenge of risking non-performing loan portfolios which eventually end up as defaulted loans. According to Ajah et al*.* (2014), the issues of loan default are lenders/borrowers related. To the lenders, there is a problem of adverse selection and to the borrower there is a problem of moral hazard with the knowledge that the loan is guarantee. Loan default affects the maximization returns and portfolio growth of microfinance. Access to credit facilities had also been reported to be limited by high rate of loan default (Enimu et al., 2017). Apart from acquiring huge expenses to recovered loans from defaulters, Ajah et al. (2013) ascertain that loan default causes a considerable reduction to loanable funds. However, since future availability of loanable fund depends considerably on the rate of previous loan repayment, there is need to explore avenues to improve the loan repayment behavior of loan beneficiaries in Calabar metropolis.

The main objective of this study was to analyze the mitigation of loan default in Nigeria through joint liability approach: The case of beneficiaries of microfinance bank agricultural loans in Calabar metropolis of Cross River State. Specifically, the study analyzed the determinants of loan default among sampled beneficiaries and compared the mean repayment amount between beneficiaries of joint liability and individual liability.

EMPIRICAL REVIEW

Using a logit regression model, Balogun and Alimi (2015) studied the factors influencing loan default and delinquency in rural credit programs in Ghana; their findings showed high probability of default when one is single. Also female borrowers rarely default. Those who borrow large sum of money may not default as those who borrow little. In the study conducted by Bassey et al*.* (2016b) on analysis of loan repayment among joint liability and individual liability beneficiaries in Akwa Ibom State Nigeria, findings showed a significant difference in loan repayment between the joint and the individual liability groups of beneficiaries. Also, while age, educational attainment, availability of surety, total income of beneficiaries and loan size were found to enhance loan repayment, household size and interest rate impacted loan repayment performance negatively. Bassey et al*.* (2016b) also studied the repayment behavior/performance between joint liability and individual liability beneficiaries in Akwa Ibom State, Nigeria, using a Z-Test analytical tool and found out that there exists a significant difference in the mean amount of loan repaid by joint liability.

MATERIALS AND METHODS

Study area

The study was conducted in Calabar, Cross River State, Nigeria. Calabar lies between latitudes 04^0 30" North of the Equator and longitudes 8 11' 21'' and 8 30'000' East of the Meridian. The town is bordered on its Eastern and Western side by two large perennial streams viz: The Great Kwa River and the Calabar River respectively. It has an area of 406 km^2 and a population of 371,022 (National Population Commission, 2006). The area is situated in the Southern geographical zone of the state which comprises Calabar Municipality and Calabar South. The main vegetation type in the study area is the mangrove forest, which gives rise to the existence of wetlands. The city of Calabar is known for her hospitality especially the Christmas Carnival which is a yearly event. The area is also blessed with fish and a lot of sea foods.

The two major seasons in Calabar are the rainy season which lasts from April to October and dry season, from November to March. Calabar has total annual precipitation that exceeds 3,000 mm annually. Temperature is respectively constant throughout the course of the year, with average temperature usually ranging from

Table 1. Microfinance banks and number of loan beneficiaries.

Microfinance banks	Total no of loan beneficiaries	Individual	Joint	Total no of beneficiaries sampled
Calabar	171	30	27	57
Ekondo	72	7	15	24
Lapo	117	24	15	39
	360	63	57	120

Source: Field Survey (2017).

25 to 28°C (Nigeria Meteorological Station, 2015). The major occupation of the people is farming and majority of them grow vegetables such as fluted pumpkin and water leaf.

Population of the study

It consists of all the registered Microfinance institutions within Calabar metropolis.

Sampling technique

Respondents used for this study were selected using the multistage sampling technique. The first stage involved a random selection of three microfinance bank: Calabar microfinance bank, Ekondo microfinance bank and Lapo microfinance bank respectively. The second stage involved the selection of thirty-nine respondents from LAPO, fifty-seven respondents from Calabar Microfinance Bank and twenty-four respondents from Ekondo Microfinance Bank making a total selection of 120 respondents (joint 57 and individual 63) that was used for the study. This selection was done in proportion to the size of registered loan beneficiaries in the selected MFIs constituting 33.3% of the total beneficiaries (Table 1).

Method of data collection

Data for the study were collected with the aid of a structured questionnaire to bring about relevant information from the respondents in the study area.

Data analysis

Different analytical techniques were used to analyze the data obtained. Both descriptive and inferential statistics were used such as mean, simple percentages, Probit model and Z statistics.

Model specification

Probit model

The Probit Model used in analyzing the determinant of loan default. It is implicitly expressed as

$$Y = bo + B_iX_i + ei \quad (1)$$

where Y = endogenous variable which takes the value of 1 if a loan beneficiary does not default and 0 otherwise.

The explicit form of the model is given as:

$$Y=bo + B_1X_1 + B_2XB_2 + B_3 X_3 + B_4 X_4 \ldots B_9 X_9 + ei \quad (2)$$

X_1 = Age of beneficiaries (Years)
X_2 = Sex of beneficiaries (male = 1, otherwise 0)
X_3 = Household size (numbers of persons in a household)
X_4 = Educational level of the beneficiaries (Years of formal education)
X_5 = Availability of surety (Yes =1, otherwise 0)
X_6 = Income of beneficiaries (naira)
X_7 = Interest amount charged on loan (naira)
X_8 = Loan amount granted (naira)
X_9= Business experience (years)
ei = Error term

These variables are similar to those of Bassey et al. (2016b).

RESULTS AND DISCUSSION

Determinants of loan default among sampled beneficiaries

The diagnostic statistic (Table 2) showed the improvement of fit made by the explanatory variables included chi-square statistic of 23.87 which was significant at 1% level of probability, implying that the regressors included in the model significantly predicted the regressand in the probit regression. Two key variables significantly influenced loan default among sampled beneficiaries. The variables were agricultural business experience and household size.

The result indicated that household size had a negative sign, and was significant at 1% level of probability. This implies that as household size decrease the probability of not defaulting will increase by 0.36% and vice versa. This is in line with *apriori* expectation because higher household sizes implied high dependable ratio. The implication is that a greater part of the loaned amount may be channeled into meeting family needs.

The variable for agricultural business experience was inversely and significantly related to loan default at 5% level. Its coefficient shows that as years of business experience decreases the probability of not defaulting increases by 0.033%. This result implies that longer experience in agribusiness, does not necessarily, lead to a better loan repayment rate. The possible reason for this could be due to better knowledge, attitude, skill and high

Table 2. Probit regression model showing determinant of loan default.

Variable	Coefficient	Standard error	Z value
Constant	3.282275	1.543824	2.12**
Age	0.0287969	0.210429	1.37
Sex	-0.0723302	0.35025393	-0.21
Educational qualification	-0.0227354	0.0742441	-0.31
Household size	-0.3561424	0.1029627	-3.46*
Annual income	-3.27e-06	2.95e-06	-1.11
Amount obtained	-9.80e-06	9/06e-06	-1.08
Business experience	-0.0331526	0.0158274	-2.09**
Log likelihood	-33.279798		
Chi square	23.87		
Pseudo R^2			

Source: Field survey Analysis (2017). * = Significant at 10%; ** = Significant at 5%; *** = Significant at 1%.

Table 3. Analysis of loan repayment performance.

Beneficiary group	Frequency	Amount obtained	Amount repaid	Rep. rate
Joint	57	8,445,000	8,039,450	95.20
Individual	63	7,030,000	6,645,526	94.53
Total	120	15,475,000	14,684,976	94.89

Source: author's Computation from Field Survey (2017).

level of education among the respondents.

Loan repayment performance by sample beneficiaries

Findings on loan repayment performance as indicated in Table 3 showed that repayment rate were 95.20 and 94.53% for joint and individual beneficiaries respectively. In Table 3, the total sum of fifteen million four hundred and seventy-five thousand naira was obtained (₦15,475,000) by beneficiaries, only fourteen million six hundred and eighty-four thousand nine hundred and seventy-six naira (₦14,684,976) representing a repayment rate of 94.89%. A breakdown of the repaid amount showed that eight million thirty-nine thousand four hundred and fifty naira (₦8,039,450) and six million six hundred and forty-five thousand five hundred and twenty-six naira (₦6,645,526) translating into 95.20 and 94.53% were repaid by joint and individual liability beneficiaries respectively. The highest repayment rate of 95.20% was recorded by the joint liability beneficiaries group and exceeded that of individual liability beneficiaries by 0.67% implying that joint liability beneficiaries had a higher repayment performance than their individual liability beneficiaries' counterpart. This finding supports other findings (Bassey et al*.,* 2016b) which reported that joint liability approach enhances loan repayment.

Comparison of mean repayment amount between beneficiaries of joint liability and individual liability

The mean comparison between repayment amount from individual and joint liability revealed that there was a significant difference in their repayment amount. Result in Table 4 shows the difference in the mean repayments amount between the loan beneficiaries. Findings showed that there was a significant difference in amount of loan repaid by the two groups of beneficiaries (joint and individual liability beneficiaries) at 99% confidence level. This is evidenced in the calculated Z value of 3.894 which was greater than the tabulated value of 1.96. This result supports the work done by Bassey et al*.* (2016).

Conclusion

The study concluded that joint liability is a better

Table 4. Z test showing mean repayment amount between beneficiaries of joint liability and individual liability.

Beneficiary group	No.	Mean amount	S.D of amount repaid	Zcal
Joint liability individual liability	57	₦134833	125581.9	3.894
	63	₦ 65317	50997.76	

Source: Data analysis (2017).

approach when it comes to borrowing than the individual liability. From the result of this study it shows that repayment rate of beneficiaries is higher when they borrow as a group and not otherwise. This is because in group lending members are able to select trust worthy peers, monitor the use of loan proceeds as well as the enforcement of repayment of the borrowed funds. Furthermore, the study showed that the key determinants of loan default were household size and business experience.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Agbaeze EK, Onuwka IO (2014). Impact of Micro credit on poverty Alleviation in Nigeria. The case study of Enugu East Local Council. International Journal of Business Management Review 2(1):27-51

Ajah EA, Eyo EO, Abang SO (2013). Repayment Performance Among Cassava and Yam Farmers under Nigerian Agricultural Bank Smallholders Loan Scheme in Cross River State, Nigeria. British Journal of Economics Management and Trade 3(4):453-467

Ajah EA, Eyo EO, Ofem UI (2014). Analysis of creditworthiness and loan repayment among bank of agriculture loan beneficiaries (poultry farmers) in Cross River State, Nigeria. International Journal of Livestock Production 5(9):155-164.

Ajah EA, Igiri JA Ekpenyong HB (2017). Determinants of Access to Credit among Rice Farmers in Biase Local Government Area of Cross River State, Nigeria. Global Journal of Agricultural Sciences 16(1):43-51.

Balogun C, Alimi RM (2015). Using repayment Data to Test Across Models of joint liabilities lending. The economic Journal 117(17):50-51.

Bassey A, Ayi S, Effiong O, Steve R (2016a). The economics of lending with joint liability: theory and practice. Journal of Development 60(7):129-131.

Bassey N, Agom D, Ikpe I (2016b). Joint liability approach a loan default mitigating strategy among beneficiaries of Micro Finance Bank loans in Akwa FFf Ibom State, Nigeria. Paper presented at the Nigerian Association of Agricultural Economists (NAAE) 17th Annual Conference. From 17th -20th October 2016 at Federal University of Technology, Owerri.

Bassey NE (2014). Determinants of loan Repayment. A study of rural women fish traders in Akwa Ibom State, Nigeria. British Journal of Economics, Management and Trade 4 (4): 541-550.

Enimu S, Eyo EO, Ajah EA (2017). Determinants of Loan Repayment among Agricultural micro finance group members in Delta State Nigeria. Journal of Financial Innovation 3(21):1-12.

Ike OA, Idoge AS (2016). Group lending repayment incentives and social collateral. Journal of development Economics 46(4):1-18.

Ike OA, Umuedafe CC (2015). Funding sources for microfinance banks in Nigeria. In the Nigerian.

Mamum F, Wahab W, Malarvizhi P, Ariapun C (2015) Banking theory and practice Vikas publishing house PVT LTD, New Delhi. Nigeria Meteorological Station (2015). Annual Report.

NPC (2006). National population commission, Nigeria. Country Wide Census Conducted in 2006.

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Evaluation of genotype × environment interaction effect on performance of garlic (*Allium sativum* L.) genotypes in Tigray region, Northern Ethiopia using AMMI and GGE biplot analysis

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Garlic is the major bulb crop next to onion in Ethiopia. Lack of stable and high yielding cultivars is one of the major problems for production and productivity of garlic in the country. Identification of adaptable, stable and high yielding genotypes under varying environmental conditions prior to release as a cultivar is the first steps for plant breeding. Therefore, developing high yielding and stable varieties is the primary objective of garlic improvement in this country. Nine garlic genotypes were evaluated to study their adaptability and stability in eight environments of Tigray region, northern Ethiopia. The experiment was carried out in randomized complete block design with three replications in four locations over two years. In this study, additive main effects and multiplicative interaction (AMMI) and genotype by environment interaction (GGE) biplot analyses were used in the evaluation of test environments and genotypes. The AMMI analysis showed that the effects of genotype, environment and genotype × environment interactions were significant (P<0.01) on bulb yield. AMMI evaluation confirmed that the three main components accounted for 89.8% of the whole genotype by environment interaction. The which-won-where view of the GGE biplot showed that environments used for this study grouped in to two mega-environments, with two different winning genotypes G9 and G7. Both AMMI and GGE biplot analysis identified promising genotypes. Genotype G9 (Bora-1/16) had the highest average yield performance and stability compared to other cultivars and should be used in breeding programs for new garlic variety development.

Key words: Additive main effects and multiplicative interaction (AMMI), bulb yield, garlic, genotype by environment interaction (GGE) biplot, stability analysis.

INTRODUCTION

Garlic (*Allium sativum* L.), is from the genus Allium and family Alliaceae grown as edible bulbous crop in the world. It is originated in Central Asia (Brewster, 1994). It is a diploid with the basic chromosome number of 2n=16

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oldest cultivated vegetable and second most extensively species of obligated apomixes, therefore its reproduction system is vegetative through its cloves. Garlic is the produced Allium next to onion (Batth et al., 2013; Diriba, 2016; Dejen, 2018). It is used for seasoning in many ingredients as well as for medicinal and spiritual purposes (Tewodros et al., 2014). It is extensively cultivated throughout the world including Ethiopia. In Ethiopia, 15381.01 ha of land have been underneath of garlic cultivation with a production of about 1386643.07 tones (CSA, 2017).

Garlic is the most widely cultivated bulbous crop in Ethiopia and it has a wide range of climatic and soil adaptation. However, its production and productivity are very low due to many biotic and abiotic bottlenecks such as lack of high yielding varieties, non-availability of quality seeds, imbalanced fertilizer use, lack of irrigation facilities, lack of appropriate disease and insect pest management and other agronomic practices, low storability, and lack of appropriate marketing services (Getachew and Asfaw, 2010; Mohammed et al., 2014). Therefore, multi environment variety trials (MET) on different crops including garlic are essential, because of the existence of genotype × environment (GE) interactions (Gauch and Zobel, 1997). The development of high yielding varieties with wide adaptability is the basic target of plant breeders. Genotype by environment interaction evaluation is important for genotype selection and cultivar recommendation, and to identify appropriate production and test environments (Singh et al., 2016; Habte et al., 2019; Ngailo et al., 2019). Bulb yield and days to maturity of garlic is disposed to environmental changes resulting in variable yield due to the significant effect of genotype-by-environment interaction (Tewodros et al., 2014).

Assessment of different genotypes across locations and over years is now not only essential to select and recommended high-yielding cultivars but also to identify suitable areas that represent the ideal environment (Yan et al., 2001). Moreover, the efficiently developed high-yielding new cultivar must have a stable overall performance and broad adaptation over a wide range of environments. A genotype is considered as stable if it has adaptability for a trait of economic significance throughout diverse environments. The environmental factor (E) usually represents the biggest issue in analyses of variance, however, it is not applicable to cultivar selection; only G and GE are relevant to significant cultivar comparison and ought to be viewed simultaneously for making selections (Yan and Kang, 2003). As there are no studies on G x E in garlic crop in Ethiopia particularly in Tigray regional state, the importance of conducting such studies throughout principal garlic producing locations have been suggested. Genotype x location (GL) interaction effects are of special interest for breeding programmes to identify adaptable, stable and high yielding genotypes and test locations. Additive main effects and multiplicative interaction (AMMI) analysis and genotype plus genotype by environment interaction (GGE) biplot analysis were widely used a multivariate technique for interaction investigation (Gauch et al., 2008; Mohammadi et al., 2010). AMMI biplot evaluation is regarded to be a high quality tool to diagnose GEI patterns graphically. AMMI analysis can also be used to find out the stability of the genotypes across locations using the (principal component axis (PCA) scores and AMMI stability value (ASV). Purchase (1997) developed the AMMI stability value primarily based on the AMMI model's principal components axis 1 and 2 scores for each cultivar, respectively.

Another powerful statistical model GGE biplot model combines the two principal effects, that is, genotypes (G) plus the G × E interaction (GE). This method is proven to be beneficial to decide which-won-where pattern of the multi-locational trials facts thereby figuring out high-yielding and stable cultivars and the power to discriminate and become aware of representative test environments (Yan, 2001). Now-a-days, it is a common practice through crop breeders to use GGE models in explaining G × E interaction and analyzing the overall performance of genotypes and test environments (Yan et al., 2007; Ngailo et al., 2019). GGE biplot, especially, is useful, to graphically represent the GE interaction, and to rank the studied genotypes and environments (Ngailo et al., 2019). According to the GGE biplot, a highly stable genotype would have a shorter projection on to the average environment coordinate (AEC) abscissa, irrespective of its direction (Yan and Kang, 2003). These two statistical analyses (AMMI and GGE) have broader relevance for agricultural researchers because they pertain to any two way data matrices, and such data emerge from many kinds of experiments (Gauch, 2006). In Ethiopia, there is no ample information on the genotype by environment interaction effects on bulb yield and yield related traits of garlic. Therefore, the objective of this study were: to assess the stability and yield performance of garlic genotypes over years and across locations; and to identify stable and high yielding candidate genotype(s) for possible release.

MATERIALS AND METHODS

Study sites and planting materials

The field experiments were conducted in four diverse garlic growing environments in Tigray region, northern Ethiopia (Ahferom, Hagere selam, Hatsebo and Ofla). The study areas represent low to high altitudes and vary in agro-ecological conditions (Table 1). The genotypes were obtained from Debre Zeit Agricultural Research Center and Axum Agricultural Research Center (Table 2). The experimental design used was a randomized complete block with three replications at each location and year. The experimental plots consisted of 6 rows of 3 m length each. Row-to-row and plant-to-plant distances were kept at 30 and 10 cm, respectively at all the locations. The genotypes were planted in the first week of

Table 1. Geographic and environmental situations of experimental areas

Location		Year	Soil type	Altitude (msl)	Rainfall average (mm)	Temperature		Geographical position	
Code	Names					Min (°C)	Max (°C)	Latitude	Longitude
E1	Hagere Selam	2017	Vertisols	2632	675	10.03	23.66	13° 38.8'N	39°10.33'E
E2	Ofla	2017	Vertisols	2133	717	10.6	24.9	12°30.8'N	39°16.54'E
E3	Hatsebo	2107	Vertisoil	2100	680	12.2	26.8	14°6′N	38°48.6'E
E4	Ahferom	2017	Cambisols	2027	715	11.15	27.32	14°16.4′N	39°3.8'E
E5	Hagere Selam	2018	Vertisols	2632	675	10.03	23.66	13°38.8'N	39°10.33'E
E6	Ofla	2018	Vertisols	2133	717	10.6	24.9	12°30.8'N	39°16.54'E
E7	Hatsebo	2018	Vertisols	2100	680	12.2	26.8	14°6′N	38°48.6'E
E8	Ahferom	2018	Cambisols	2027	715	11.15	27.32	14°16.4′N	39°3.8'E

Table 2. List of the garlic genotypes used for the study cropping season.

Genotype label	Name of Genotype	Origin/Source
G1	Briki-Gc-1/16	AxARC
G2	Kuriftu(S.check)	DZARC
G3	birki-Gc-2/16	AxARC
G4	Bora-Gc-16	AxARC
G5	Bisheftu Nech(S.check)	DZARC
G6	Tseday(S.check)	DZARC
G7	Bora-2/16	AxARC
G8	Bora-3/16	AxARC
G9	Bora-1/16	AxARC

AxARC= Axum Agricultural Research Center, DzARC = Debre Zeit Agricultural Research Center

November for two consecutive years (2017 to 2018/2019) under irrigation. Di-ammonium phosphate (DAP) as a source of phosphorus was applied at the rate of 200 kg ha^{-1} during planting and nitrogen fertilizer was applied in the form of Urea at the rate of 150 kg ha^{-1} in splits, half during transplanting and the rest as side dressing at 45 days after transplanting. Furrow irrigation method, scheduled at 8-12 days interval (AxARC, 2016) was used. Weeding and other management practice have been accomplished as required for each site. Data were recorded on 90% physiological maturity, plant height, bulb diameter, bulb weight, number of cloves per bulb and bulb yield per hectare. The yield harvested from four central row of each net harvestable plot in kg was once transformed into tha^{-1}.

Statistical analysis

The analysis of variance was carried out for each location over two years using SAS version 9.2 (SAS, 2008) and before combining the data, the assumption of (ANOVA) normality test was executed the for bulb yield. Mean comparison was executed using LSD at 5 and 1% level of significance. Genotype -by- environment interaction impact that was detected in ANOVA table that led to the GEI and stability analysis to be done using AMMI and GGE biplot (Yan, 2001). AMMI analysis was performed following the AMMI model in accordance to Gauch (2013) using R software model 3.4.4. The AMMI stability values (ASV) were calculated as advised via Dagnachew et al. (2014). GGE biplot analysis, on the other hand, was used to carry out the usage of the genotype via environment analysis in R software v3.4.4 (Yan et al., 2000; R Team, 2018; Habte et al., 2019). Thus, the first two principal components (PC1 and PC2) were used to graphically represent the GEI, to become aware of the rank of studied genotypes and environments (Yan et al., 2000). The AMMI statistical model is given below:

$$Y_{ijk} = \mu + g_i + e_j + \sum_{k=1}^{n} \lambda_k a_{ik} \gamma_{ik} + \varepsilon_{ij}$$

Where: Y_{ijk} the yield of the i^{th} genotype in the j^{th} environment, μ the grand mean, g_i the mean of i^{th} genotype minus the grand mean, ej the mean of j^{th} environment minus the grand mean, λ_k is the square root of the eigen value of the principal component analysis (PCA) axis, a_{ik} and γ_{jk} are the principal component scores for PCA axis n of the i^{th} genotype and j^{th} environment and εij is the error. According to Zobel et al. (1988), AMMI with only two interaction principal component axes could be the best predictive model. Hence, two IPCAs were adopted in this study in AMMI analysis. AMMI stability value (ASV) was calculated to quantify and rank of genotypes. This was carried out using the formula below which is suggested by Purchase (1997). The AMMI stability value (ASV) described by Purchase et al. (2000):

$$ASV = \sqrt{\left[\frac{SSIPCA1\ sum\ of\ square}{SSIPCA2\ sum\ of\ square}(IPCA1\ Score)\right]^2 + (IPCA1\ scpre)^2}$$

Table 3. Mean squares of combined analysis of variance of bulb yield of nine garlic genotypes evaluated across locations in (2017-2019).

		Mean squares					
Sources	**DF**	**PH**	**BD**	**BW**	**NCPB**	**DTM**	**BYLD**
Genotype(G)	8	117.96**	15.56ns	50.62**	287.62**	1141.46**	17.48**
Location(L)	3	2344.5**	843.59**	875.33**	348.02**	3946.19**	22.95**
Season(S)	1	1081.2**	1183.48**	1562.89**	363.17**	18481.5**	216.36**
GXL	24	16.97ns	18.24**	18.31ns	16.21**	64.95**	2.59**
GXS	8	15.77ns	28.34**	30.25*	11.91*	16.74ns	2.89**
LXS	3	1359.73**	489.96**	817.96**	284.03**	6817.94**	168.79**
GXLXS	24	14.81ns	11.64ns	15.42	5.63ns	54.476**	2.64**
Error	136	15.87	8.98	14.48	5.8	17.26	1.028
CV (%)		7.4	7.4	13.55	15.72	3.16	15.26
R^2 (%)		87	84	81	88	96	89
Mean		53.80	40.57	28.1	15.37	131.5	6.64

Ns, * and ** = non-significant, significant and highly significant at 0.05 and 0.01level of significance respectively. DF= degree of freedom, PH= plant height, BL= blub length, BD= bulb diameter, BW= bulb weight, NCPB= number of cloves per bulb, DTM= days to maturity and BYLD= bulb yield (tha^{-1}), R^2 (%) = coefficient of determination and CV = coefficient of variation.

Where: $\frac{IPCA1\ sum\ of\ square}{IPCA2\ sum\ of\ square} =$ represents the weighted value assigned to the first interaction principal component score due to its high contributions in the GE model,SSIPCA1 and SSIPCA2 are the sum squares of for IPCA1 and IPCA2 ,respectively. And also IPCA1 and IPCA2 are the first and second IPCA scores for each genotype. The larger ASV the more specifically adapted the genotype is to a certain environment and the smaller ASV indicates a more stable genotype across environments (Purchase, 1997; Ngailo et al., 2019). The GGE biplot were constructed from the first two principal components (PC1 and PC2) derived by subjecting the environment centered yield data (which contains G and GE) to singular valued composition (SVD) (Yan, 2000; Yan et al., 2007). Based on singular value decomposition of the first two principal components, is:

$$Yij - \mu - aj = e1\ b1\ cj1 + e2\ b2\ cj2 + \varepsilon ij$$

Where, Yij is the measured mean of genotype i in environment j, µ is the grand mean, aj is the main effect of environment j, i +aj is the mean yield across all genotypes in environment j, e1 and e2 are the singular values for the first and second principal components, respectively b1 and b2 are eigenvectors of genotype i for the first and second principal components, respectively, cj1 and cj2 are eigenvectors of environment j for the first and second principal components, respectively, εij is the residual associated with genotype i in environment j. AMMI and GGE biplot were performed using R software program Version 3.4.4.

Yield stability index (YSI) and Rank-Sum (RS)

The new approaches known as YSI and RS were calculated by the following formulas:

YSI = RASV + RY

Where RASV is the rank of AMMI stability value and RY is the rank of mean bulb yield of genotypes (RY) across environments. YSI incorporate both mean yield and stability in a single criterion. Low value of this parameter shows desirable genotypes with high mean yield and stability. Rank sum (RS) = Rank mean (R) + Standard deviation of rank (SDR). RS incorporate both yield and yield stability in a single non-parametric index. Genotypes with the least RS are considered stable with high bulb yield under irrigated conditions. Standard deviation of rank (SDR) was measured as:

$$S_i^2 = \frac{\sum_{j=1}^{m} (R_{ij} - \overline{R}_i)^2}{l-1}$$

Where R_{ij} is the rank of X_{ij} within the j_{th} environment, Ri. (R), is the mean rank across all environments for the ith genotype and SDR= $(S^2i)^{0.5}$.

RESULTS AND DISCUSSION

Analysis of variance

The combined analysis of variance (Table 3) for bulb yield and yield related traits showed highly significance differences (P≤0.01) among genotypes, locations and presence of significance G×E interaction, indicating variation of environments and the presence of genetic variability among genotypes. The analysis of variance also indicated that there was highly significant variation (P<0.01) among the genotypes in plant height (cm), bulb diameter (mm), bulb weight (g), number of cloves per bulb, days to 90% physiological maturity and bulb yield (tha^{-1}). This showed variability among the genotypes for these characters. The overall mean bulb yield of the locations was ranged from 5.58 to 8.39 tha^{-1} (Table 4), therefore, the eight environments (Location × year) showed broad variation in their yield production potential. The mean bulb yield of genotypes was 6.64 tha^{-1} (Tables

Table 4. Overall means for bulb yield (tha^{-1}) and other yield related characters of garlic genotypes grown across locations during 2017-2019.

Genotypes code	Genotype	PH	BD	BW	NCPB	DTM	BYLD
G1	Briki-Gc-1/16	55.12^{ab}	40.4a^{bc}	26.79^{d}	15.9^{c}	136.2^{bc}	6.36^{cd}
G2	Kuriftu	53.9^{b}	40.57^{bc}	26.42^{d}	16.1^{c}	136.12^{bc}	6.06^{de}
G3	birki-Gc-2/16	54.77^{b}	40.16^{bc}	27.64^{bcd}	16.6^{c}	137.29^{ab}	6.55^{cd}
G4	Bora-Gc-16	51.13^{c}	39.2^{c}	27.85^{bcd}	10.5^{d}	126^{e}	6.19^{cd}
G5	Bisheftu Nech	54.26^{b}	40.2^{bc}	26.99^{cd}	16.8^{c}	134.54^{c}	5.58^{e}
G6	Tseday	57.32^{a}	40.9^{ab}	29.57^{ab}	20.8^{a}	139.29^{a}	6.68^{c}
G7	Bora-2/16	55.44^{ab}	41.96^{a}	29.09^{abc}	18.5^{b}	129.33^{d}	7.61^{b}
G8	Bora-3/16	51.33^{c}	40.2^{bc}	27.6^{bcd}	11.8^{d}	126.2^{e}	6.37^{cd}
G9	Bora-1/16	50.97^{c}	41.45^{ab}	30.83^{a}	11.3^{d}	118.46^{e}	8.39^{a}
	LSD (0.05)	2.27	1.71	2.17	1.38	2.37	0.58
	CV (%)	7.4	7.4	13.55	15.72	3.16	15.27
	Mean	53.80	40.57	28.1	15.37	131.5	6.64

Means with the same letter are not significantly different. BYLD= bulb yield (tha^{-1}), BD=bulb diameter (mm), BW= bulb weight (g), NCPB=number of cloves per bulb and DTM= day to maturity.

Table 5. Analysis of variance for Additive Main effect and Multiplicative Interaction (AMMI) model of bulb yield of garlic genotypes grown at Tigray Region, northern Ethiopia (2017-2019).

Source of variation	DF	Sum of square	Mean square	% Explained
Genotype	8	14721	1840.1***	11.86
Rep(Env't)	16	2917	182.3*	
Environment	7	76770	10967.2***	61.84
Interactions(GEI)	56	14300	255.4***	11.52
IPCA1	14	7872.64	562.33***	55.1
IPCA2	12	2336.18	219.34**	18.4
IPCA3	10	1106.69	138.33**	16.3
Error	128	13289	103.8	
Total	215	124140	577.4	

GEI= Genotype by Environment interaction; DF= Degrees of freedom.

3 and 4). The performance of genotypes in all yield associated characteristics was different both in each and throughout locations. This indicated that the efficiency of a breeding program aimed only at yield enhancement resulted in inconsistent performance of genotypes due to significant genotype- by- environment interaction effect, which complicates the procedure of crop variety development particularly when varieties are chosen in one environment and used in others (Singh et al., 2016).

AMMI analysis of variance for bulb yield

Combined analysis of variance for bulb yield of the nine garlic genotypes examined across eight environments is presented in Table 5. The main effect differences among genotypes, environments, and the interaction results were highly significant (P < 0.01) of the whole variance of bulb yield. The environment impact accounted for 61.84%, whereas genotype and G × E interaction results accounted for 11.86 and 11.52% of the total variation, respectively (Table 5). The maximum environmental sum square indicated that there was a huge difference among the testing locations causing unique genotypes to perform in another way across the testing environments and the excessive percentage of the environment is an indication that the main factor that influence yield performance of garlic genotypes in Ethiopia is the environment. Genotypes revealed highly significant (P<0.001) variations for bulb yield. This shows that there was genetic difference among genotypes for this trait. This variation is beneficial when intending to find out about the consequences of G×E interaction, as properly as to consider the phenotypic stability of genotypes. The magnitude of the GEI sum of squares used to be rather similar with that of the genotypes, indicating that there

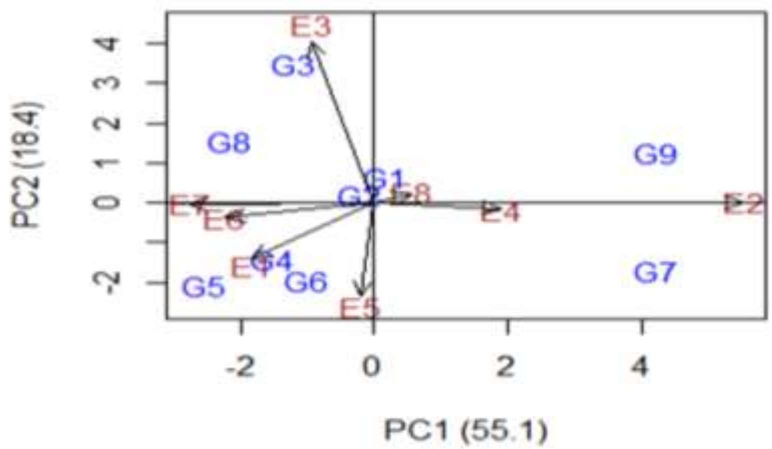

Figure 1. AMMI biplot of nine genotypes of garlic evaluated across locations for their bulb yield.

was by some means comparable response of some of the genotype across environments.

The results of AMMI model for bulb yield are presented in Table 5. As it is viewed from Table 5, the mean square of the three IPCA was highly significant ($P<0.001$). The AMMI biplot, which accounted for 73.5% of the G×E interaction, gives the interaction principal component rankings of the 1st and 2nd IPCA. The first PC axis (PC1) score explained 55.1% of the variant in GEI, while the 2nd PC axes accounted for18.4% of the variability. Many researchers witnessed that the best accurate AMMI model prediction can be made by using the first two IPCAs (Yan et al., 2000). Therefore, the dataset obtained from the interaction of 9 genotypes examined at eight environments were well predicted through the first two IPCAs. On the other hand, the IPCA scores of a genotype in the AMMI analysis are indication of the stability of a genotype throughout environments (Purchase, 1997). Accordingly, the closer the IPCA scores to zero (origin), the more stabile the genotypes are across all environments (Purchase, 1997). The IPCA1 used to be plotted on x-axis whereas IPCA2 was plotted on y-axis for bulb yield and yield components (Figure 1). The greater the IPCA scores (positive or negative) as it is a relative value, the greater especially adapted a genotype is to certain environments. The greater IPCA scores approximate to zero, the more stability the genotype is throughout environments sampled (Purchase, 1997). The IPCA1 and IPCA2 rankings of bulb yield for each genotype and the corresponding AMMI stability value (ASV) are presented in Table 6. According to ASV ranking, genotype 2 had the lowest value indicating its high stability, while genotypes 9 and 7 were extraordinarily unstable. Purchase (1997) pointed out that the closer the genotypes score to the center of the biplot the genotype is broadly adapted and the reverse is true. Regarding the position of the environments in the biplot graph, locations E2 (Ofla) and E4 (Ahferom) were the most discriminating environments as they have long distance between their marker and the biplot origin (Figure 1). However, due to their massive IPCA2 score, genotypic variations discovered at these environments may not precisely show the genotypes average yield across locations. The interaction principal component one (IPCA1) and the interaction principal component two (IPCA2) scores in the AMMI model are indications of stability.

Considering the first interaction principal component (IPCA1), the genotype G5, was the most stable genotype with IPCA1 value (-2.53). When the second interaction principal component (IPCA2) was considered, G5 was the most stable genotype with interaction principal component value (-2.07) followed by the genotype G6 with the IPCA2 value (-1.94). The two principal components have their own extremis, however calculating the AMMI ASV is a balanced measure of stability (Purchase, 1997). The genotype with lower ASV values is viewed stable and genotypes with higher ASV are unstable. Based on the value of ASV, genotype G2 was the most stable with an ASV value of 0.67 followed with the genotypes G1 and G6 with ASV value of 0.84 and 3.59 of bulb yield respectively. Genotypes G7, G9 and G5 were the most unstable with ASV value of 12.71, 12.64 and 7.86 of bulb yield respectively (Table 6).

Yield stability index (YSI)

Stability is not the only criterion for selection, due to the fact that most stable genotypes would no longer necessarily provide the best yield across environments (Mohammadi et al., 2010), consequently there is a need for approaches that comprise both mean yield and stability in a single index number of authors introduced extraordinary standards for simultaneous selection of yield and stability rank-sum, modified rank-sum and yield

Table 6. Mean bulb yield (tha^{-1}) of garlic varieties evaluated at eight environments in Tigray Region, northern Ethiopia.

Code	Environment													
	E1	E2	E3	E4	E5	E6	E7	E8	Mean	ASV	IPCA1	IPCA2	YSI	RS
G1	6.56	10.16	6.96	5.82	6.22	3.41	6.30	5.47	6.36	0.71	0.18	0.62	8	8.9
G2	6.09	9.77	6.46	6.07	6.03	2.91	7.04	4.06	6.06	0.42	-0.21	0.19	9	10.1
G3	5.88	9.28	8.46	6.99	5.82	3.21	7.91	4.90	6.56	4.06	-1.19	3.49	10	6.0
G4	6.30	8.72	5.76	7.33	6.36	3.18	8.02	3.87	6.19	2.96	-1.51	-1.41	11	9.9
G5	6.46	8.66	5.44	3.89	6.67	3.32	7.17	3.11	5.58	4.84	-2.53	-2.07	16	12.1
G6	7.01	9.51	6.35	6.79	7.66	3.70	7.39	5.33	6.78	2.6	-1.01	-1.94	6	5.9
G7	6.63	13.64	6.53	9.05	7.89	3.72	7.84	6.14	7.68	7.48	4.21	-1.72	11	4.8
G8	6.04	9.43	7.42	5.57	6.01	4.16	8.28	4.09	6.37	4.01	-2.13	1.54	10	8.1
G9	7.54	14.97	9.18	8.54	8.49	4.41	8.09	6.29	8.44	7.38	4.21	1.26	9	3.9

G1 to G9 name of genotypes; E1= Hagere Selam in 2017, E2= Ofla in 2017, E3= Hatsebo in 2017, E4= Ahferom in 2017, E5= Hagere Selam in 2018, E6= Ofla in 2018, E7=Hatsebo in 2018, and E8= Ahferom in 2018. IPCA = Interaction Principal Component Axis, ASV = AMMI Stability Value, YSI=Yield stability Index, RS= rank sum.

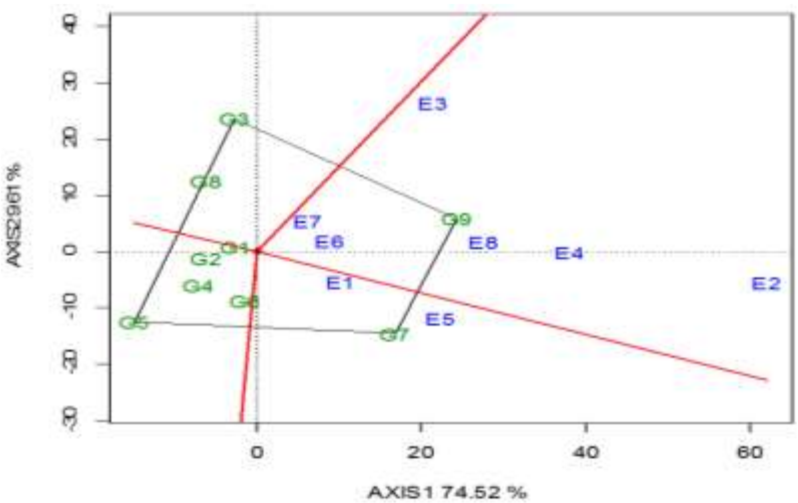

Figure 2. The which-won-where view of the GGE biplot to show which genotypes performed bests in which environments representing bulb yield of nine garlic genotypes (G) evaluated in eight environments (E) in 2017 and 2018/2019.

stability (Farshadfar et al., 2011). In this regard, ASV takes into account both IPCA1 and IPCA2 and justifies most of the variation in the GEI. The genotype with least YSI is regarded as the most stable with high yield mean. It was utilized to identify high yielding and stable genotypes in cereal crops like durum wheat (Mohammadi et al., 2010). Based on YSI, the most stable genotype with high bulb yield is genotype G9 with YSI of 9 accompanied via G7 and G6 YSI of 11 and 11, respectively .The highest YSI indicate that G4, G5 and G8 were unstable genotypes. Rank-sum (RS) showed that genotype G9 produced high bulb yield and followed via genotype G7 indicating that they were the most stable genotypes with high bulb yield. Both YSI and RS confirmed that genotype G7 gave high bulb yield.

Analysis of GGE biplot for bulb yield

GGE Biplot analysis shows “which-won-where” pattern, ranking of cultivars on the basis of yield and stability, and correlation vectors among environments. Angles between environment vectors were used to judge correlations (similarities/dissimilarities) between pairs of environments (Yan and Kang, 2003; Yan, 2011). GGE biplot is visualized on the basis of consequences defined for the first two principal components (Yan et al., 2001). In the current study, the first two principal components of GGE biplot explained 84.13% (PC1=74.52 and PC2=9.61%) of the whole variations (Figure 2). In the polygon, genotypes located far away from the origin are the vertex genotypes having the highest yield in the region (Esayas et al.,

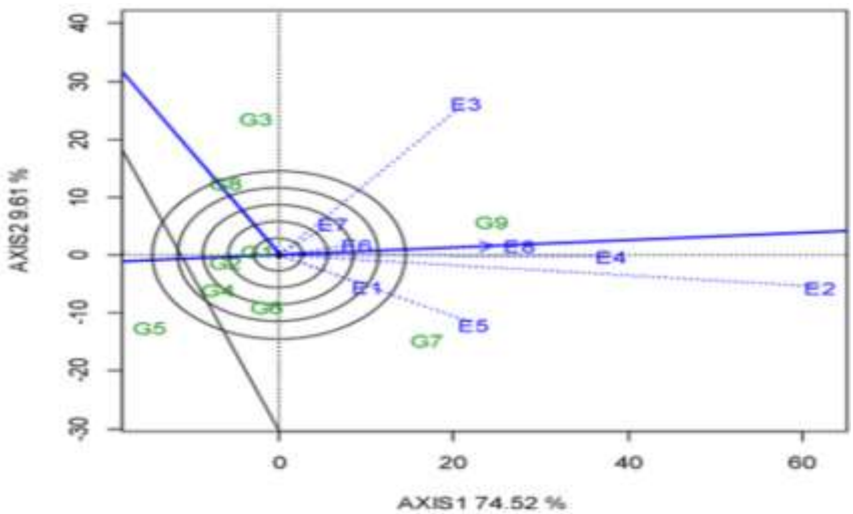

Figure 3. GGE biplot view showing the relationship among the testing environments and discriminativeness vs representativeness.

2019; Habte et al., 2019). In this study, genotypes G9, G7, G5 and G3 had the highest yield in their respective sector.

The GGE Biplot graphic analyses of the nine garlic genotypes tested at eight environments are presented in Figure 2. Rays in Figure 2 divided the biplot into four sectors. The environments were located in two mega-environments where group 1 contained environments E2,E3,E4, E6, E7 and E8 and group 2 had two environments E1 and E5 (Figure 2), while the genotypes were located in all four sectors. The genotypes found at vertex of the sectors are the most profitable genotypes of that sector (Yan and Tinker, 2006).The genotype on the vertex of the polygon, contained in a mega-environment, had the highest yield in at least one environment and was one of the best-performing genotypes in the other environments (Yan, 2002). Accordingly, the vertex genotype G9 (Bora-1/16) was high yielder in most of environments except E1 and E5 where G7 (Bora-2/16) was the winner. Therefore, genotype G9 was best yielder in environments E2, E3, E4, E6, E7 and E8. Genotype 7 gave highest yield in E1 and E5 (Figure 2 and Table 6). The other vertex genotypes (G3 and G5) were not the highest yielding genotypes at any environment. Thus genotypes G9, G7, G3 and G5 are specifically adapted. G1 and G2 genotypes were closest to the center of origin; therefore they were broadly adapted genotypes.

Relationship among environments and discriminative vs. representativeness

The vector view of a GGE biplot provides a summary of the interrelationships among the environments (Yan, 2002). Provided that the biplot explained an adequate amount (≥50%) of the total variation, the correlation coefficient between any two environments is reliable (Yan et al., 2000). Furthermore, the length of an environmental vector is an estimation of discriminating power of the environment (Yan et al., 2007). Accordingly, the results of the present study revealed that the first principal component (PC1) and the second (PC2) respectively clarified 74.52% and 9.61% of the variance (Figure 3). The two principal component axis (PC1 and PC2) together clarified 84.13% of the total variance. So this biplot can be used for extracting interrelationships among the environments.

A long environmental vector represents a high capacity to discriminate the genotypes. With the longest vectors from the origin, environment E2 was the most discriminating of the genotypes, while E3, E4, E5 and E8 were moderately discriminating. However, with the shortest vector from the origin, E7 provided little or no information about the genotype differences. Furthermore, the vector view of the GGE-biplot provides a brief summary of the interrelationships among the environments. Two environments are positively correlated if the angle between their vectors is <90°, negatively correlated if the angle is >90°, independent if the angle is 90° (Yan and Tinker, 2006). Based on this, E2, E4, E6, E7 and E8 environments were positively correlated because all of the angles among their vectors were smaller than 90°. However, the angle between vectors of tester E3 andE1, E3 and E5 were approximately 90°, and were not correlated (Figure 3).

Ranking testing environments relative to the ideal environment and genotype

Average environmental axis (AEA) is a line passing via the origin and pointing to the positive direction with its

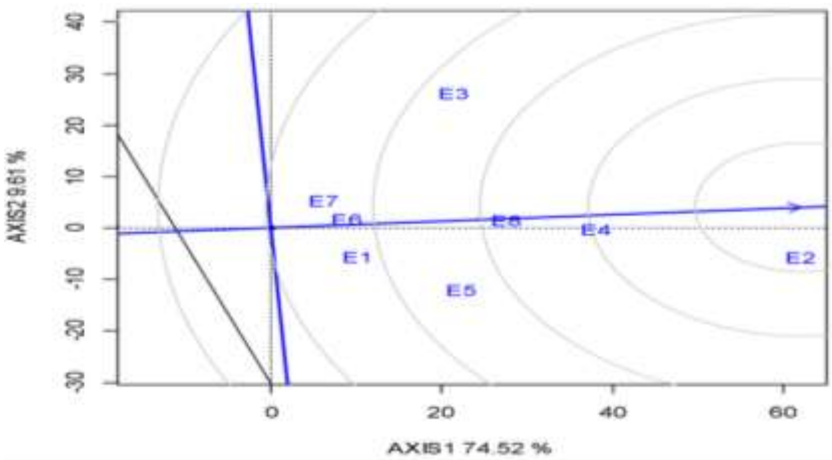

Figure 4. GGE biplot showing ranking of test environments relative to an ideal test environment.

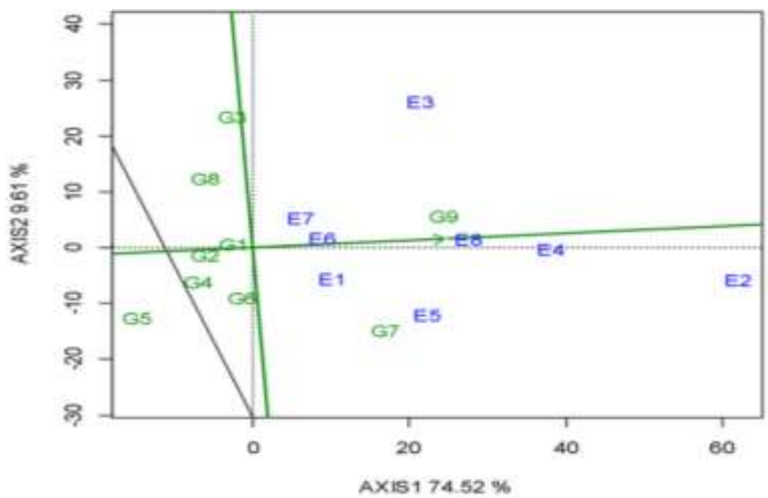

Figure 5. GGE biplot graph based on genotype-focused scaling for comparison of genotypes with ideal genotype.

distance equal to the longest vector. An ideal environment is representative and has the highest discriminating power (Yan and Tinker, 2006). The ideal environment is located in the first concentric circle in the environment-focused the GGE biplot and the environments that are close to the ideal environment are defined as the desired environments. Based on this, E2 located in the first concentric circle and has been the most ideal environment (Figure 4). Thus, genotype evaluation in E6 environment maximized the observed genotypic variation among genotypes for bulb yield of the tested garlic genotypes. E4 and E8 environments were close to the ideal environment (E2) and these environments were identified as suitable environments. This difference between environments can be related to soil fertility, climate changes and other environmental variations from year to year. The most acceptable is the one closest in the sketch of the ideal environment (Yan et al., 2000). The environments E2 (Ofla, 2017), E4 (Ahferom, 2017) and E8 (Ahferom, 2018) contained in the third concentric circle is the place with best potential to discriminant genotypes, favoring the choice of ideal genotypes (Figure 4 and Table 6).

Ranking genotypes relative to the ideal genotype and environment

An ideal genotype is defined as the one with the highest yield across the test environments and is definitely stable in performance (Yan and Kang, 2003). The average environment coordination view of the GGE biplot suggests the rating of genotypes primarily based on the overall performance of best genotypes (Figure 5). The

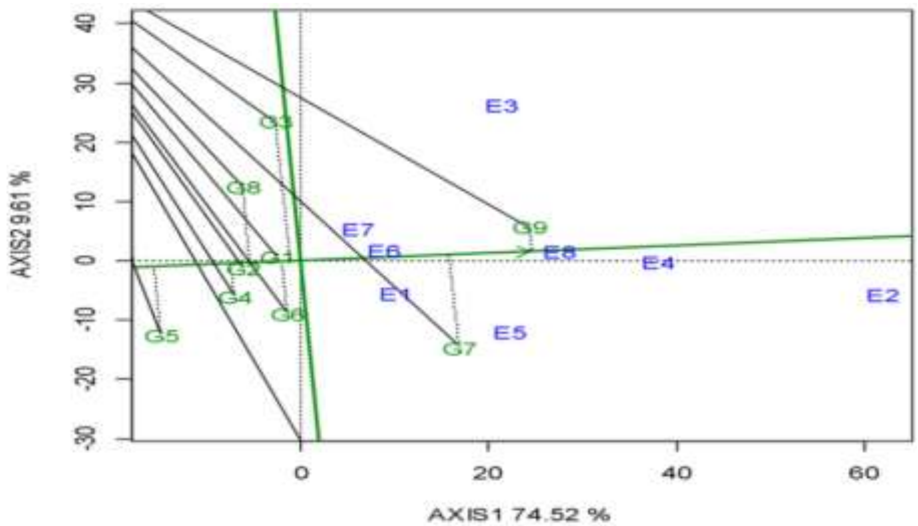

Figure 6. Average environment coordination (AEC) views of the GGE biplot ranking based on mean performance and stability of nine garlic genotypes across eight environments (E1-E8).

relative adaptation of the best genotype is evaluated through drawing a line passing via the biplot origin and the best genotype marker. This line is referred to as a genotype axis and is related to the economic profitable genotype (Habte et al., 2019). Such ranking of genotypes revealed that both G9 and G7 are the high yielding genotypes.

Genotypes mean yield vs. stability

The mean yield performance and stability of genotypes was evaluated by an average environment coordination (AEC) method (Yan, 2001, 2002). In the AEC system, AEC X axis (PC1) passes through the biplot origin with an arrow indicating the positive end of the axis and indicates the mean performance of genotypes. The ATC Y-axis passes through the biplot origin and is perpendicular to the ATC X-axis. This axis indicates the stability axis (PC2) (Figure 6). Based on these, statistically, the stable genotypes located near the AEC X axis (PC1) with PC2 scores of almost zero. According to Figure 6, genotypes with above average yield were G9 and G7 and located on the right side of the biplot origin, while genotypes with blow average yield were G3, G5 and G8 and located on left side of the biplot origin. A best genotype for a particular environment has the best possible mean yield and responds best at that unique environment while it is less stable in the other environments and wants to be proposed for a specific environment (Yan et al., 2001). According to the similar authors, best cultivars have large PC1 rankings (high mean yield) and small PC2 scores (high stability). Thus, in the present study, G9 and G7 which had higher PC1 and smaller PC2 rankings had been recognized as high yielding and stable. Therefore, the genotypes G7 and G9 with stable and high yield can be recommended as commercial variety for the Tigray region and others which have similar agro-ecology.

Conclusion

From the current study, it is concluded that multiple methods were employed to analyze stability. The AMMI and GGE biplot methods can be effectively utilized for the identification of the suitable genotypes for suitable environments. The results of AMMI analyses indicated that garlic bulb yield performances was highly affected by environmental effect followed by the magnitude of GEI, but genotype contributed the minimum effect. The AMMI and GGE biplot analysis permitted estimation of interaction effect of a genotype in each environment and they helped to identify genotypes best suited for specific environments. The GGE biplot analysis shown that the genotypes G9, G7, G5 and G3 were corner genotypes and suited to specific environments. The polygon views of the GGE biplot grouped in two possible mega environments. The first mega environment consisted of six environments (E2, E3, E4, E6, E7 and E8); and the second mega environment consisted of two environments (E1 and E5). In addition, the discriminating power vs. representativeness view of the GGE biplot has been an effective tool for test environments evaluation. Environment E2 (Ofla) and E4 (Ahferom) were the most discriminating for bulb yield of the tested garlic genotypes. According to the AMMI and GGE biplot models, considering simultaneous average yield and stability, G9 (Bora-1/16) and G7 were the best genotypes. Therefore, these genotypes should be released for Tigray region and others similar agro-ecologies to enhance the productivity of garlic.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Batth GS, Kumar H, Gupta V, Brar PS (2013). GGE Biplot Analysis for Characterization of Garlic (*Allium sativum* L.) Germplasm Based on Agro-Morphological Traits. International Journal of Plant Breeding 7(2):106-110.

Brewster JL (1994). Onions and other Vegetable Alliums. CABI publishing Wellesbourne, UK.

CSA (Central Statistical Agency). (2017). Agricultural sample survey Report on area and production of major crops of private peasant holdings. The FDRE Statistical bulletin 1:584. Addis Ababa, Ethiopia 118p.

Dagnachew L, Masresha F, Santie de V, Kassahun T (2014). Additive Main Effects and Multiplicative Interactions (AMMI) and genotype by environment interaction (GGE) biplot analyses aid selection of high yielding and adapted finger millet varieties. Journal of Applied Biosciences 76(1):6291-6303.

Dejen B (2018). Review on the Application of Biotechnology in Garlic (Allium Sativum) Improvement. International Journal of Research Studies in Agricultural Sciences 4(11):23-33.

Diriba-Shiferaw G (2016). Review of management strategies of constraints in garlic (*Allium sativum* L.) production. Journal of Agricultural Sciences -Sri Lanka 11(3).

Esayas T, Frehiwot G, Hussein M, Melaku T, Diribu T, Abebech S, (2019). Genotype× environment interaction by AMMI and GGE biplot analysis for sugar yield in three crop cycles of sugarcane (Saccharum officinirum L.) clones in Ethiopia. Cogent Food and Agriculture 5:1-14.

Farshadfar E, Mahmodi N, Yaghotipoor A (2011). AMMI stability value and simultaneous estimation of yield and yield stability in bread wheat (*Triticum aestivum* L.). Australian Journal of Crop Science. 5(13):1837-1844.

Gauch HG, Piepho, HP, Annicchiaricoc P (2008). Statistical analysis of yield trials by AMMI and GGE. Further considerations. Crop Science 48:866-889.

Gauch HG (2013). A simple protocol for AMMI analysis of yield trials. Crop Science 53:1860-1869.

Gauch HG (2006). Statistical analysis of yield trials by AMMI and GGE. Crop Science 46:1488-1500.

Gauch HG, Zobel RW (1997). Identifying MegaEnvironments and Targeting Genotypes. Crop Science 37:311.

Getachew T, Asfaw Z (2010). Achievements in shallot and garlic research. Report No.36. Ethiopian Agricultural Research Organization, Addis Ababa Ethiopia.

Habte J, Kebebew A, Kassahun T, Kifle D, Zerihun T (2019). Genotype-by-Environment Interaction and Stability Analysis in Grain Yield of Improved Tef (Eragrostis tef) Varieties Evaluated in Ethiopia. Journal of Experimental Agriculture International 35(5):1-13.

Mohammadi R, Mozaffar RM, Yousef A, Mostafa A, Amri A (2010). Relationships of phenotypic stability measures for genotypes of three cereal crops. Canadian Journal of Plant Science 90:819-830

Mohammed A, Shiberu T, Thangavel S (2014). White rot (*Scelerotium cipivorum Berk*)-an aggressive pest of onion and garlic in Ethiopia: an overview. Journal of Agricultural Biotechnology and Sustainable Development 6(1):6-5.

Ngailo S, Shimelis H, Sibiya J, Mtunda K, Mashilo J (2019). Genotype-by-environment interaction of newly-developed sweet potato genotypes for storage root yield, yield-related traits and resistance to sweet potato virus disease. Heliyon, Elsevier 5(3):1-23.

Purchase JL, Hatting H, Van Deventer CS (2000). Genotype × environment interaction of winter wheat (T. aestivum) in South Africa: Stability analysis of yield performance. South African Journal of Plant Soil 17(3):101-107.

Purchase JL (1997). Parametric analysis to describe genotype x environment interaction and yield stability in winter wheat, University of Free State. Bloemfontein, South Africa

R Team (2018) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna. available at http://www.R-project.org.

SAS Institute Inc. (2008). SAS/STAT ®9.2 User's Guide. Cary, NC: SAS Institute Inc

Singh RK, Dubey BK, Gupta RP (2016). Genotype × environment interaction and stability analysis for yield and its attributes in garlic (*Allium sativum* L.). Journal of Spices and Aromatic Crops 25(2):175-181.

Tewodros B, Fikreyohannes G, Nigussie D, Mulatua H (2014). Registration of 'Chelenko I'Garlic (*Allium sativum* L.) Variety Haramaya University, Ethiopia. East African Journal of Sciences 8(1):71-74.

Yan W (2001). GGE biplot: A windows application for graphical analysis of multi environment trial data and other types of two-way data. Agronomy Journal 93:1111-1118.

Yan W (2002). Singular value partitioning in biplot analysis of multi-environment trial data. Agronomy Journal 94:990-996.

Yan W (2011). GGE Biplot vs. AMMI Graphs for Genotype-by-Environment Data Analysis. Journal of the Indian Society of Agricultural Statistics 65(2):181-193.

Yan W, Tinker NA (2006). Biplot analysis of multi environment trial data: Principles and applications. Canadian Journal of Plant Science 86:623-645

Yan W, Kang MS (2003). GGE Biplot analysis: a graphical tool for breeders, geneticists, and agronomists. Boca Raton, FL: CRC Press.

Yan W, Cornelius PL, Crossa J, Hunt LA (2001). GGE Biplot analysis of multi-environment trial data. Crop Science 41(3):656-663.

Yan W, Hunt LA, Sheng Q, Szlavnics Z (2000). Cultivar evaluation and mega environment investigation based on the GGE biplot. Crop Science 40:597-605.

Yan W, Kang MS, Ma B, Wood S, Cornelius PL (2007). GGE biplot vs. AMMI analysis of genotype-by environment data. Crop Science 47:643-655.

Yan W, Wu HX (2008). Application of GGE biplot analysis to evaluate genotype (G), environment (E), and G× E interaction on Pinus radiata: A case study. New Zealand Journal of Forestry Science 38(1):132-142.

Zobel RW, Wright MJ, Gauch HG (1988). Statistical analysis of a yield trial. Agronomy Journal 80:38.

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Full Length Research Paper

Yield increase of soybean inoculated with a commercial arbuscular mycorrhizal inoculant in Brazil

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Arbuscular mycorrhizal fungi (AMF) play an important role in plant growth. However, there are no records of the use of AMF-based inoculants in agricultural crops in Brazil. The objective of this work was to evaluate the agronomic efficiency of a commercial inoculant containing the AMF *Rhizophagus intraradices* in combination with phosphate fertilization in soybean under different edaphoclimatic conditions in Brazil. Experiments were conducted in five states (Goiás, Mato Grosso, Minas Gerais, Paraná and Rio Grande do Sul) in a 2 x 3 factorial scheme, with two inoculation treatments (inoculated and non-inoculated seeds) and three doses of phosphate fertilization (0, 50 and 100% of the recommendation). At the end of the crop cycle (stages R2), it was found that the inoculant provided average increases of 29% in biomass (regardless of the applied P dose) and grain yield, and higher P uptake. It is concluded that the inoculant increases biomass production, P uptake and soybean yield under different edaphoclimatic conditions in Brazil, especially in soils that originally had low or medium levels of available P on the ground.

Key words: Arbuscular mycorrhizae, soybean, phosphate fertilization, *Rhizophagus intraradices*.

INTRODUCTION

Arbuscular mycorrhizal fungi (AMF) are obligate biotrophic fungi that establish symbiotic relationships with plant roots (Smith and Read, 2008). The association provides benefits for plant growth and enables the AMF to complete their life cycle (Smith and Read, 2008). AMF absorb water and nutrients from the soil and transfer them to plants, while plants provide photoassimilates to fungi (Bi et al., 2005). The mycelium formed by the AMF outside the roots allows the plant to explore a larger volume of soil, and this extension of the root system increases the intake of water and nutrients (Smith and Read, 2008).

The symbiotic relationship between fungi and plants provides a number of benefits to surrounding plants and the environment (Jeffries et al., 2003; Berruti et al., 2016). Phosphorus (P) supply is the most important feature mediated by AMF (Smith and Smith, 2011). This benefit is ultimately observed primarily due to the low mobility of P in the soil (Smith and Read, 2008). AMF can contribute to plant growth through increased drought

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tolerance (Garg and Chandel, 2010), increased supply of other nutrients such as nitrogen, potassium and calcium (Sharif et al., 2011; Dania et al., 2013; Crespo, 2015). They also contribute to the formation of soil aggregates by the production and release of glycoproteins such as glomalin (Rillig, 2004). Arbuscular mycorrhizal fungi interfere with plant growth and soil biology the same way chemistry and physics influence the establishment of symbiosis between plants and AMF. The availability of P in soil is linked to the production of signals and responses between plants and AMF species, directly related to soil characteristics (Smith et al., 2011; Cely et al., 2016).

Around 300 species of AMF have been formally described (Glomeromycota Species List, 2019). Among those, *Rhizophagus intraradices* (N.C. Schenck & G.S. Sm.) C. Walker & A. Schüßler (until recently classified as *Glomus intraradices*) is a generalist species found distributed throughout different environments. The list of plant species benefited by AMF on growth and productivity include crops of agricultural importance such as soybeans (Spagnoletti and Lavado, 2015), corn (Guo et al., 2014), beans (Tajini et al., 2012), wheat (Ardakani et al., 2011) and cotton (Orak and Demir, 2011). Because of these characteristics, the use of that species for the formulation of commercial inoculants is recommended.

The production of AMF inoculants bumps into the obligatory symbiotic character of these fungi. This makes it difficult to develop an efficient inoculant production system that delivers large volumes of propagules in a short period. Traditional models of multiplication of these fungi use trap plants or root tissue culture (Vosátka et al., 2012; Berruti et al., 2015). Records of an efficient production process capable of supplying the Brazilian agricultural market with arbuscular mycorrhizal inoculants have not been found in the literature.

Farmers have increased their interest in sustainable practices, and this includes introducing, maintaining, and increasing AMF populations in production systems. The application of AMF in agriculture in annual crops, which represent large areas of agricultural production in Brazil (IBGE, 2019) should be studied. Thus, considering that the arbuscular mycorrhizal species *R. intraradices* has the potential to assure high yields in agricultural production systems with more sustainable management, studies are needed to prove these effects.

Thus, the objective of this work was to evaluate the agronomic efficiency of a commercial inoculant based on the AMF *R. intraradices*, in combination with different levels of phosphate fertilization, on soybean (*Glycine max* L.) growth and yield in five locations with different edaphoclimatic conditions in Brazil.

MATERIALS AND METHODS

Experiment locations

The experiments were conducted in the 2016/2017 crop season (September 2016 to March 2017), considering the soybean planting window, cultivars and driving practices of each tested site. For this, five (5) representative sites of soybean cultivated areas in the country (Figure 1) and with distinct edaphoclimatic characteristics were selected: Padre Bernardo in Goiás (GO), Ritápolis in Minas Gerais (MG), Tangará da Serra in Mato Grosso (MT), Pitangueiras in Paraná (PR), and Cachoeira do Sul in Rio Grande do Sul (RS) (Table 1).

Experimental specifications

Aiming to validate the efficiency of the AMF-based inoculant, the experiments were conducted in the field, following the protocols defined by MAPA (Brazilian Ministry of Agriculture, Livestock and Supply) for product registration based on the plant growth-promoting microorganism specifications, specifically the regulations contained in the IN SDA 13, from 03/25/2011 (Brasil, 2011) and the IN SDA 53, from 10/24/2013 (Brasil, 2013). Among the main requirements for the registration of microbial inoculants, we highlight the need to prove agronomic efficiency in at least four (4) regions with distinct edaphoclimatic characteristics, and that the role of the inoculant in reducing fertilization for the crop is proven.

All fertilization procedures, based on soil analysis and expected yield (SBCS, 2004), and crop treatments (Table 2) followed the recommendations for each site. The experiments were conducted until the complete pre-maturation cycle of the grains was completed, that is, in the R7 stage (Oliveira et al., 2016) (Table 1).

Experimental set-up

The experiments were implemented in subdivided plots, with mechanized sowing. In all cases, the experiments followed a 2 x 3 factorial scheme, with two inoculation levels (inoculated and non-inoculated seeds), three (3) levels of phosphate fertilization (no fertilization, 50%, and 100% of the recommendation for each site), and six replications (6), in subdivided blocks.

Every one of the 36 plots of each experiment had an area of 24 m^2 (4 m x 6 m), with a spacing of 1 m between plots. Each plot had a working area of 10 m^2, formed by the central portion of each plot (6 lines with a spacing of 0.5 m x 4 linear meters).

Inoculant characteristics

Rootella BR® (later registered under No. 22902 10000-0) was obtained by a mixed fungal propagule production system using trap plants (Berruti et al., 2015) and tissue culture (Diop, 2003; Srinivasan et al., 2014; Schuessler, 2015).

The inoculant was characterized and its purity certified, noting the number of propagules and the exclusive presence of propagules of the AMF species *R. intraradices*. This approach considered characteristics inherent to fungal spores, such as number, thickness, coloration, and ornamentation of the layers that cover the spore (https://invam.wvu.edu/). For inoculant quantification, the Most Probable Number (MPN) method (Oblinger and Koburger, 1975) was adopted to verify the concentration of 2,500 propagules per gram of product, as indicated on the label. The inoculant tested contained a mixed composition of spores and other fungal propagules (hyphae) and was formulated using sterile ultrafine vermiculite as an inert component.

Crop management

The seeds were inoculated with the initial addition of a liquid adhesive (Symbiosis Pró - MAPA - RS - 12834-1) and incorporation of the inoculant immediately before sowing. The recommended use

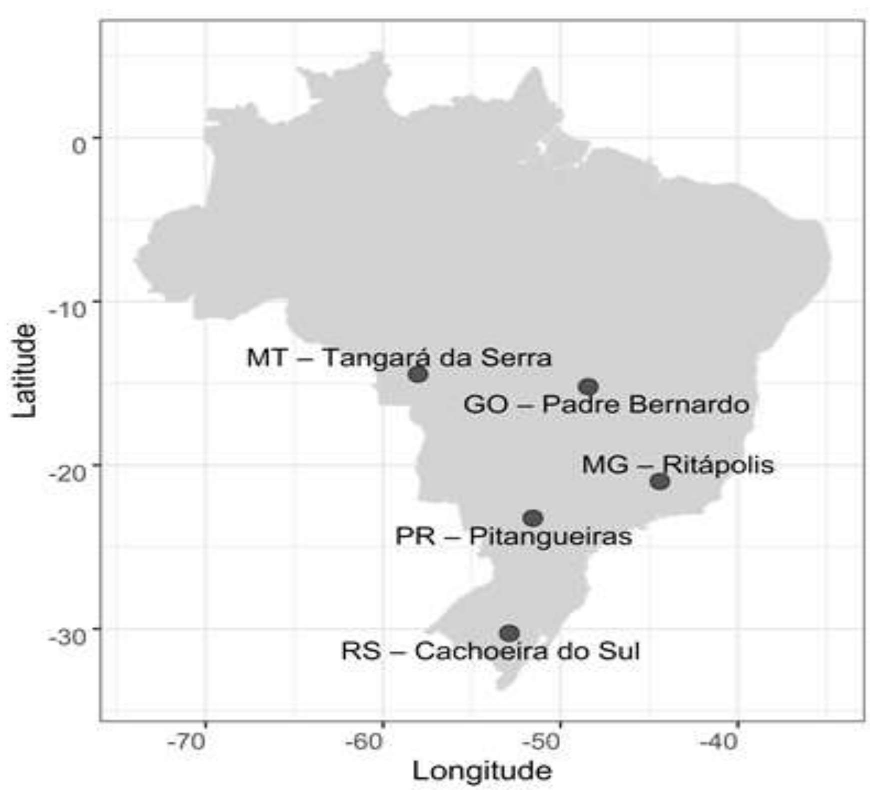

Figure 1. Locations of municipalities where experiments were conducted.

Table 1. Information on planting, harvesting, cultivar, soil characteristics, chemicals and agronomic practices employed in each location.

State/ municipality	Planting date (2016)	Harvesting date (2017)	Plant density (working area)#	Cultivar	Soil characteristics		
					P (mg/dm^3)†*	Clay (g kg^{-1})	N-P_2O_5-K_2O (kg ha^{-1})
GO – Padre Bernardo (15°12'29.9"S 48°26'28.6"W)	Nov 23	Mar 07	144	M8372 IPRO	6.71 (M)	600	0-80-40
MG – Ritápolis (20°59'39.1"S 44°24'15.4"W)	Oct 26	Mar 09	149	Nidera NS 7000 IPRO	1.10 (VL)	230	0-120-120
MT – Tangará da Serra (14°26'43.7"S 58°02'14.4"W)	Nov 07	Feb 15	288	TMG 2185 IPRO	4.80 (M)	680	0-120-120
PR – Pitangueiras (23°14'34.7"S 51°33'30.3"W)	Sep 21	Feb 27	224	DM 6563 IPRO	12.33 (H)	400	0-95-100
RS – Cachoeira do Sul (30°16'35.7"S 52°53'07.4"W)	Sep 04	Mar 01	151	Nidera 4823	16.00 (H)	260	0-60-140

† P determined by Mehlich 1 and the classes determined according to fertilizer recommendation system for each location; # Evaluations were based on the same number of plants per working area (30 plants); * Levels of P in the soil. M: Medium; VL: Very low; H: High.

Table 2. Edaphoclimatic characteristics and fungicides applied in each location.

State	Soil type	pH (H_2O)	Average temperature (°C)	Precipitation (mm/year)	Fungicides applied via seed	Fungicides applied during plant growth
GO	RL[1]	5.3	22.9	1431	Maxim XL[†]	Fox[‡] + Elatus*
MG	SCL[2]	5.3	19.2	1456	Maxim XL	No application
MT	RYL[3]	5.6	24.8	1830	Maxim XL	Fox + Cypress** + Priori Xtra[#]
RS	RU[4]	5.1	19.4	1692	Standak top (Estrobilurina)	Fox
PR	RL	5.9	19.0	1500	Standak top (Estrobilurina)	No application

[1] Red Latosol; [2]Sandy Clay Loam; [3]Red-yellow Latosol; [4]Red Udult. [†] Maxim XL: Fludioxonil + Metalaxyl−M; [‡] Fox: Trifloxystrobin + Protioconazol; *Elatus: Azoxystrobin **+** Benzovindiflupyr; **Cypress (Ciproconazole + difenoconazole); [#]Priori Xtra (Ciproconazole + azoxystrobin).

for the formulation tested was 1.0 kg of inoculant per hectare, regardless of the plant stand used. The plants were also inoculated via seed at the time of planting with nitrogen-fixing bacteria following the recommendations for soybean cultivation in the country.

Crop treatments performed in each area and region are described in Table 2. For one of the evaluated states (RS), seeds were treated with specific fungicides before planting. In addition to seed treatment, fungicides were also used throughout crop development in the state of RS, as specified in Table 2. The choice of products was based on the history of fungal diseases in each evaluated site. Weed and insect control was carried out using specific herbicides and insecticides for each situation.

Evaluations

Shoot dry biomass yield, P biomass content and accumulation, and grain yield was evaluated as required by MAPA IN SDA 13 and IN SDA 53. At the phenological stage R7 (Oliveira et al., 2016), 50 plants were collected from the four central lines of each plot (working area of 10 m^2). Drying of the biomass was performed in a forced air circulation oven at 60 °C to constant weight. Grain yield was determined from the yield obtained for 30 plants evaluated per plot at harvest time (considering the area mean stand), which were oven-dried following the same procedure described above.

The contents and accumulations of P in the tissues were determined from a sample composed of 20 newly mature leaves (without the petioles) harvested from the working area of each plot at the end of flowering (phenological stage R2) (Oliveira et al., 2016). Determinations followed Tedesco et al. (1995). Data was tested for homoscedasticity (Bartlet) ($p \leq 0.05$). Two-way analysis of variance was performed and Tukey's test was used for mean separation at 5 and 10% probabilities, as established by the legislation for registration of inoculants (Brasil, 2011, 2013). The analysis was performed using the software SISVAR v.5.3.

RESULTS

Table 3 shows the effects of mycorrhizal inoculation, phosphate fertilization and the interaction of these factors on biomass and grain yield, along with the levels and foliar accumulation of P. In general, the mycorrhizal inoculation and phosphate fertilization significantly influenced the afore-mentioned variables, from which the main effects are presented and discussed below.

There was an increase in biomass yield linked to the increase in phosphorus supplied by fertilization in three (3) of the five (5) sites tested. In Ritápolis (MG), a site with low soil available P, the response to soybean phosphate fertilization was clearer (Figure 2e).

Inoculation provided a 29% average increase in soybean aerial biomass yield, with a range of 17 to 41% increase. At all phosphate fertilization levels, the biomass of the inoculated plants was equal to or higher than that of the non-inoculated plants. Inoculation increased biomass yield in three (3) of the five (3) areas with 0% P (average 33% increase), in all areas in the treatment with 50% of the recommended P (average 32%), and in four (4) of the five (5) areas with 100% of the recommended phosphate fertilization (average 25%) (Figure 2).

The average P contents (Figure 3) in plant tissues at the five (5) sites were 1.13 and 1.70 $g.kg^{-1}$ in the non-inoculated and inoculated plants, respectively. The highest mean content was observed in MT (2.33 $g.kg^{-1}$) (Figure 3a), and the lowest value in GO (0.79 $g.kg^{-1}$) (Figure 3d), two sites with medium P content available in the soil. In four (4) of the five (5) areas there were positive responses of P content to inoculation, except in Pitangueiras, in the State of PR (Figure 3b). The average increase coming from the inoculation on the biomass P content compared to the non-inoculated plants was 72%. In MG and RS, sites with very low and very high levels of available soil P, respectively, positive responses to inoculation were seen, regardless of the applied P dose (Figure 3e and 3c), with average increases of 84% for MG and 190% for RS.

P accumulation (Figure 4) was higher in response to inoculation in all sites with no phosphate fertilization (average increase of 155%), in four (4) of the five (5) sites with 50% of the recommended dose (average increase of 297%), and in four (4) of the five (5) sites with 100% of the recommended dose of P (average increase of 92%).

Grain yield varied in response to AMF inoculation. Grain yield was on average 29% higher in the inoculated *versus* the non-inoculated plants, reaching 65% increases in some locations (Figure 5). In MT, the average yield increase of the inoculated plants was 28% (Figure 5a). It was 46% for PR (Figure 5b), 16% for RS (Figure 5c), 15% for GO (Figure 5d), and 40% for the

Table 3. F values and significance for the variables inoculation (I), phosphate fertilization (P) and IxP interaction on biomass and grain yield, as well as phosphorus levels and accumulations in soybean in different regions of Brazil.

Location	F value			
	Dry biomass	**Leaf P content**	**P accumulation**	**Grain yield**
Tangará da Serra-MT	I = 9.2**	I = 3.6^{ns}	I = 20.6**	I = 12.4**
	P = 4.7**	P = 2.7**	P = 7.4**	P = 5.5**
	IxP = 2.9*	IxP = 1.3^{ns}	IxP = 2.3^{ns}	IxP = 2.4^{ns}
Pitangueiras-PR	I = 72.6**	I = 0.3^{ns}	I = 27.2**	I = 80.3**
	P = 7.0**	P = 0.9^{ns}	P = 4.5**	P = 11.9**
	IxP = 2.2^{ns}	IxP = 1.2^{ns}	IxP = 0.7^{ns}	IxP = 2.9*
Cachoeira do Sul-RS	I = 42.9**	I = 538.7**	I = 97.8**	I = 13.9**
	P = 12.2**	P = 5.3**	P = 3.5**	P = 9.4**
	IxP = 2.4^{ns}	IxP = 5.1**	IxP = 4.8**	IxP = 1.6^{ns}
Padre Bernardo-GO	I = 19.7**	I = 4.8*	I = 17.1**	I = 6.3*
	P = 2.4^{ns}	P = 2.9*	P = 5.4**	P = 1.2^{ns}
	IxP = 1.1^{ns}	IxP = 2.1^{ns}	IxP = 0.7^{ns}	IxP = 0.4^{ns}
Ritápolis-MG	I = 22.1**	I = 41.8**	I = 73.3**	I = 7.5**
	P = 176.3**	P = 7.3**	P = 53.2**	P = 74.4**
	IxP = 3.4*	IxP = 0.9^{ns}	IxP = 6.3**	IxP = 2.4^{ns}

I=Inoculation; P=Phosphate fertilization; IxP= inoculation and phosphate interaction; **, * Significant effect at 5 and 10% probability; ns = non-significant effect.

state of MG (Figure 5e). Inoculation provided grain yield increases at two (2) of the five (5) sites where no phosphate fertilization was applied (average increase of 34%), in three (3) of the five (5) areas with 50% of the recommended dose of P (average increase of 35%), and in four (4) of the five (5) areas with 100% of the recommended P (average increase of 22%) (Figure 5). Grain yield responded to different rates of P in three of the tested areas, with higher intensity in Ritápolis (MG) (Figure 5e), the site with the lowest initial P content available in the soil.

Comparing the average yields of the three P levels with the non-inoculated treatment at 100% of the recommended P dose, inoculation provided grain yield increases of up to 29% (Figure 5). Grain yield of the inoculated plants was equal to or greater than the non-inoculated plants that received 100% of the P dose recommended. This was true to almost all sites and P dose treatments, the only exception being the 0% P inoculated treatment in MG, a site originally with low P available in the soil.

DISCUSSION

In most situations, there was a positive response to the application of the tested AMF inoculant. The literature provides substantial data on the beneficial effects of *R. intraradices*. This species is considered generalist, not only being associated with soybeans, but also with many other plant species of agronomic interest. Among the species that are known to benefit from interaction with *R. intraradices* are maize, beans, wheat, rice, oats and barley (Guo et al., 2014; Spagnoletti and Lavado, 2015; Tajini et al., 2012; Ardakani et al., 2011). Species of the genus *Rhizophagus* (formerly classified as *Glomus*) are cosmopolitan, and are present practically worldwide (Davison et al., 2015), and like other microorganisms such as *Rhizobium* and *Bradyrhizobium*, generally bring remarkable benefits when associated with plant species.

The effects of AMF on plant association may vary with the environmental conditions to which plants are exposed, such as higher or lower P availability in the soil. As observed in the present study (Figure 2), Cely et al. (2016), in a study conducted in the State of Paraná, showed that plants fertilized and inoculated with *Rhizophagus clarus*, a physiological species very close to *R. intraradices*, showed biomass increases of 24% when compared to fertilized and non-inoculated plants. This increase is very similar to that observed in the present work, with an average of 25% in fertilized (100% P) and inoculated plants. Thus, it can be stated that the inoculation promoted higher biomass yield, due to the greater efficiency of the inoculant, approaching the yields

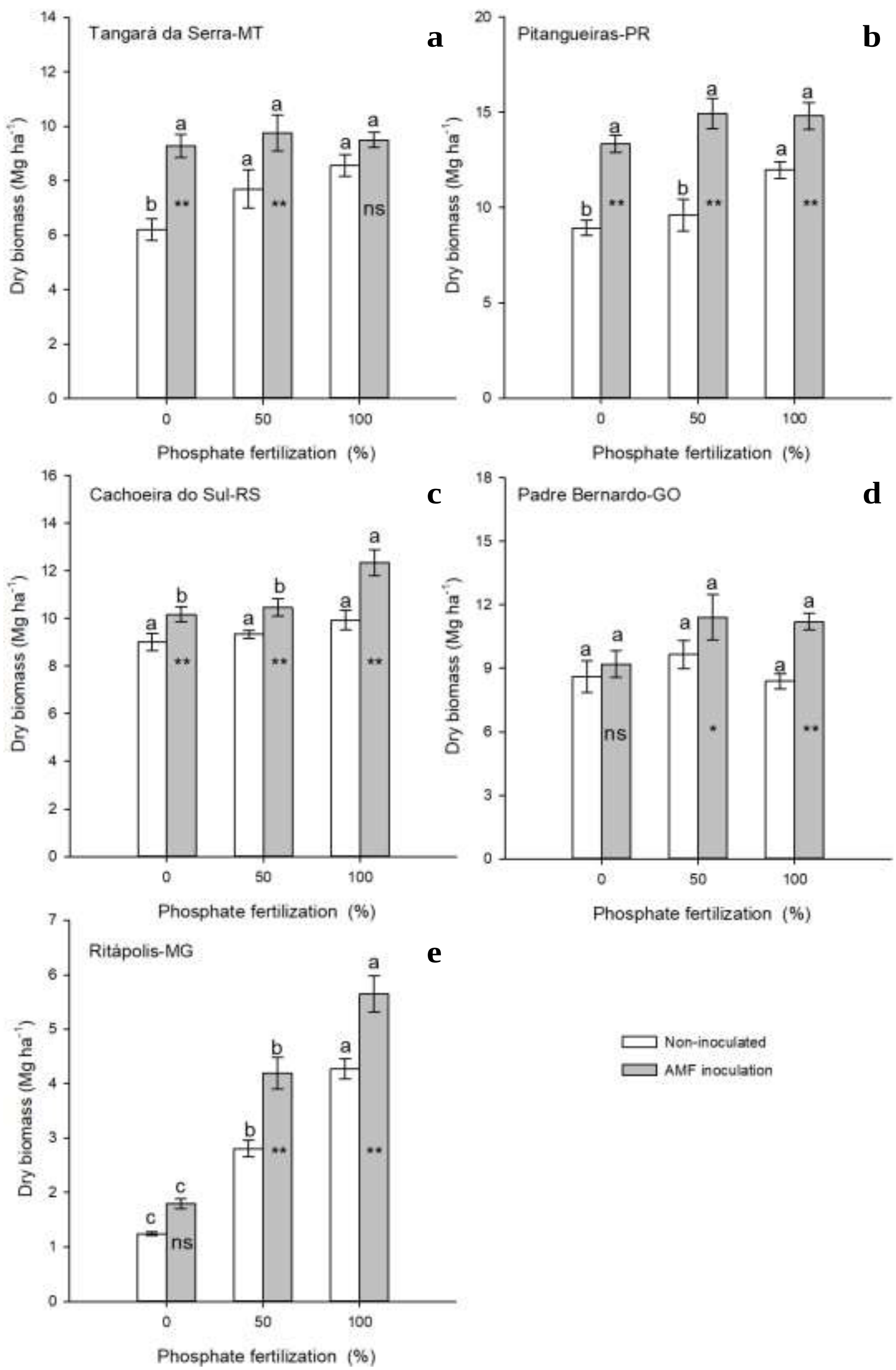

Figure 2. Effect of mycorrhizal inoculant application and phosphate fertilization on dry biomass of soybean (Mg ha^{-1}). **, * Significant effect of inoculation at 5 and 10% probability by F test within P doses; ns = non-significant effect of inoculation. Vertical bars represent the standard error. Averages followed by the same letter within the same inoculation treatment do not differ by Tukey's test at 5%/10% probability.

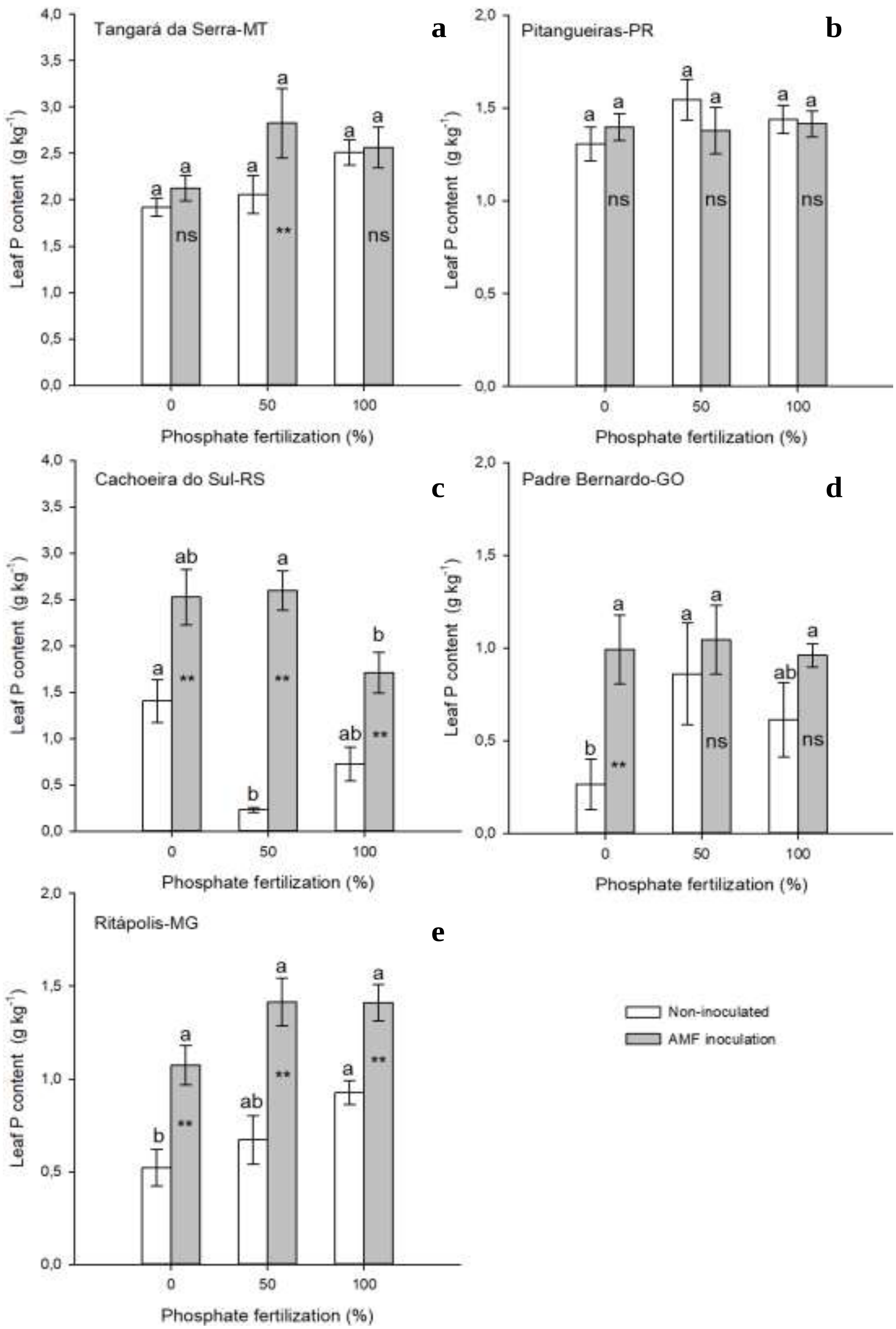

Figure 3. Effect of mycorrhizal inoculant application and phosphate fertilization on the level (g kg^{-1}) of foliar P on soybean. ** Significant effect of inoculation at 5% probability by F test within P doses; ns = non-significant effect of inoculation. Vertical bars represent the standard error. Averages followed by the same letter within the same inoculation treatment do not differ by Tukey's test at 5%/10% probability.

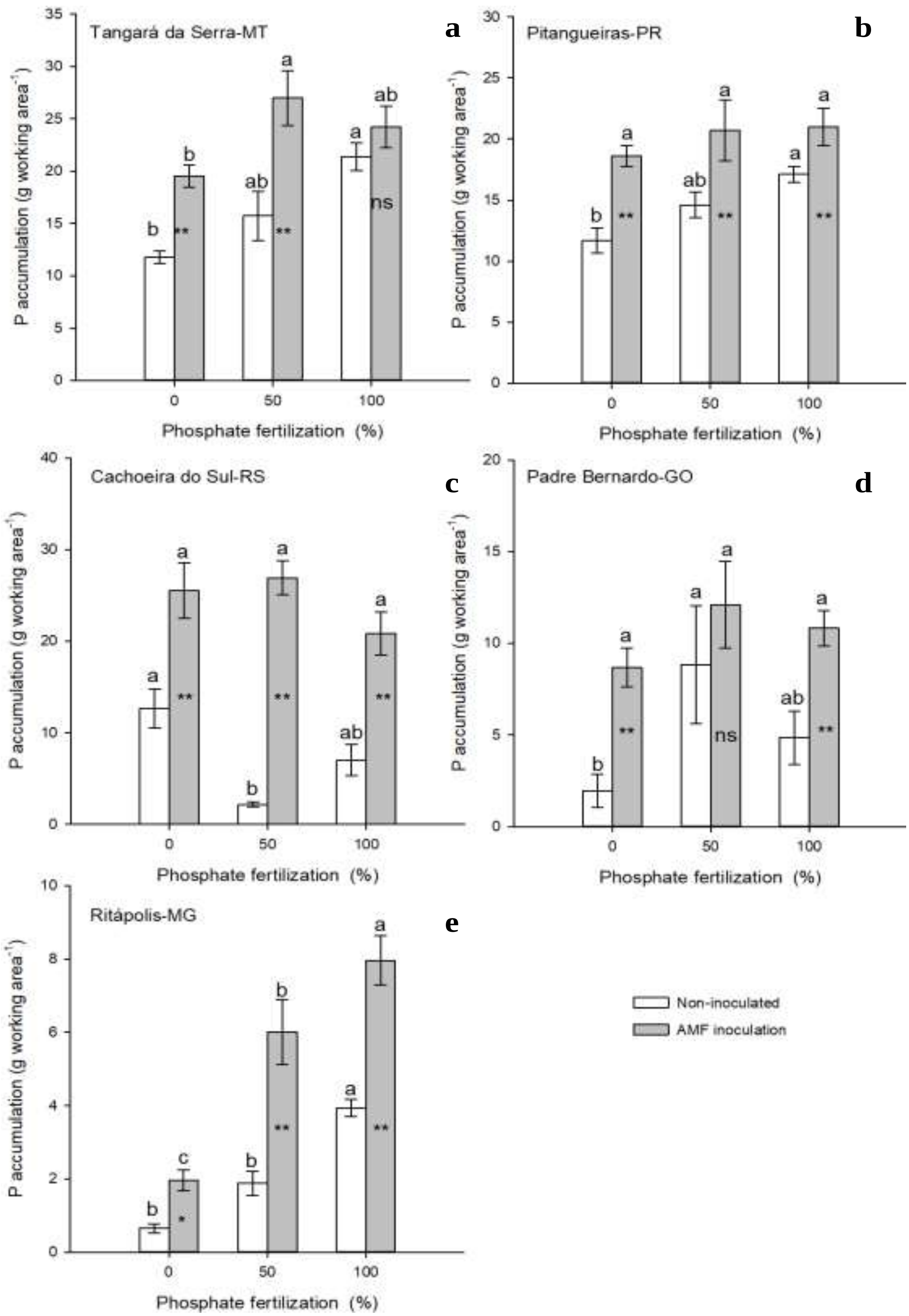

Figure 4. Effect of mycorrhizal inoculant application and phosphate fertilization on the accumulation (g working area^{-1}) of foliar P on soybean. **, * Significant effect of inoculation at 5 and 10% probability by F test within P doses; ns = non-significant effect of inoculation. Vertical bars represent the standard error. Averages followed by the same letter within the same inoculation treatment do not differ by Tukey's test at 5%/10% probability.

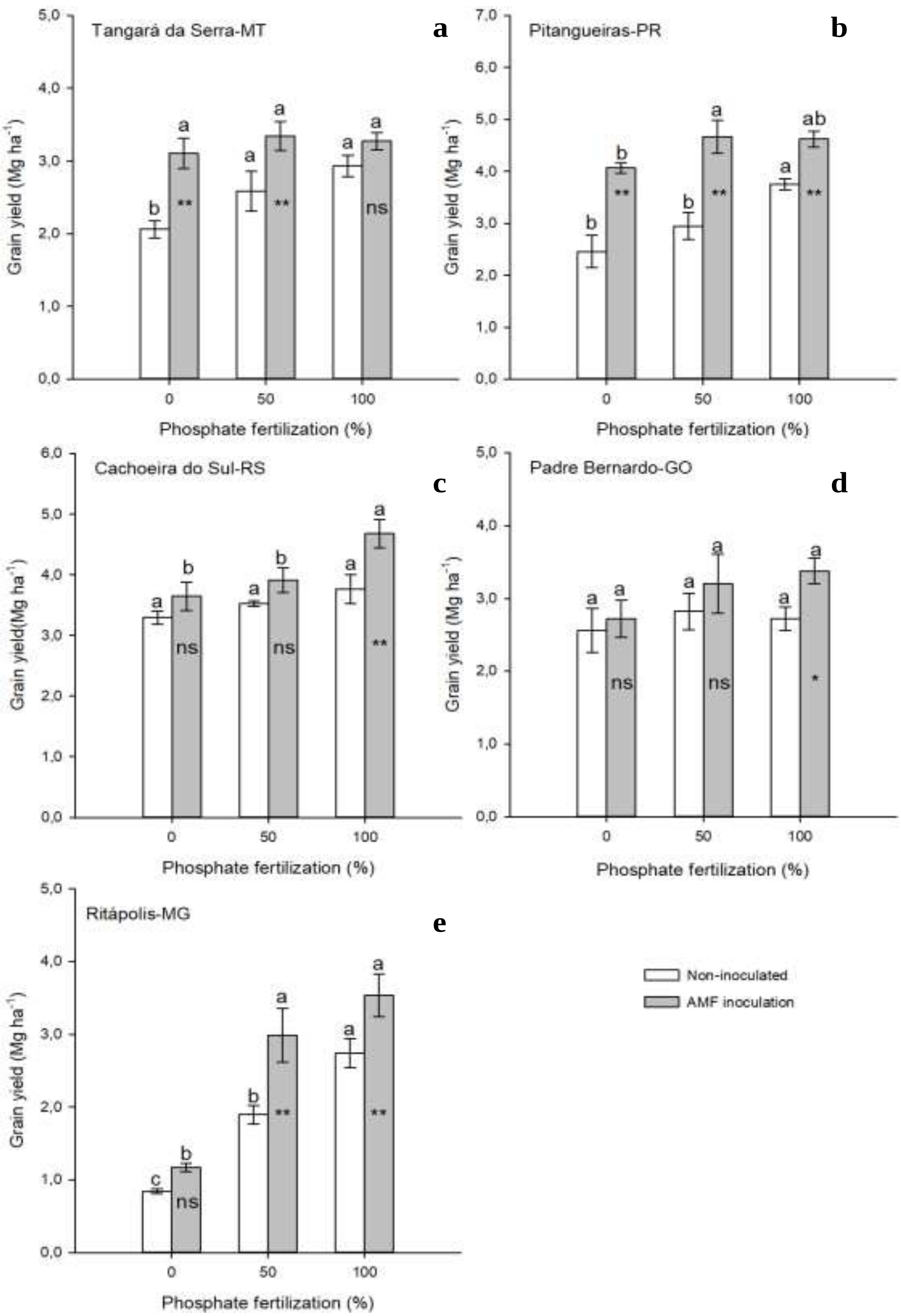

Figure 5. Effect of mycorrhizal inoculant application and phosphate fertilization on grain yield of soybean (Mg ha^{-1}). **, * Significant effect of inoculation at 5 and 10% probability by F test within P doses; ns = non-significant effect of inoculation. Vertical bars represent the standard error. Averages followed by the same letter within the same inoculation treatment do not differ by Tukey's test at 5%/10% probability.

observed for grain.

For normal plant growth during the growing season, Marschner (1995) shows that the P content should be between 3.0 and 5.0 g P kg^{-1} in the tissues, being toxic when exceeding 10 g P kg^{-1}. Some studies with soybean varieties have observed average levels that fall within this range (Corrêa et al., 2004; Broch and Ranno, 2012). However, in the present work, the average levels did not exceed 2.33 g of P kg^{-1} in the dry biomass. Rezende et al. (2009) also observed values lower than those described by Marschner (1995), with minimum and maximum contents of 0.97 to 1.23 g kg^{-1} of P. Thus, it is believed that although there are tables in the literature describing ranges for P values for soybean, these values may vary depending on the cultivation conditions and soybean cultivars evaluated.

In MG, a site with low available P, the effect of the inoculation on P content was clear (Figure 3e), supplying the P demand under low availability conditions. However, in places with higher P availability, such as in the State of RS, there seems to be a stimulus for extra P consumption. Even in the plots that did not receive phosphate fertilization, the P content is higher than in the control treatment (non-inoculated with 100% of P). The scientific literature highlights the potential of plant association with AMF in soils with different P concentrations available on plant nutrition and development (Harley, 1989; Bolan, 1991; George et al., 1995; Harrison, 1995; Bago et al., 2002; Ohtomo and Saito, 2005; Hodge et al., 2010; Smith et al., 2011; Cely et al., 2016). Thus, it can be seen that plants established in soils with high P content may respond differently to AMF inoculation compared to plants from low soil P available.

The largest increases in P accumulation in biomass were in RS (Figure 4c). This may have been caused by concentration as a function of higher P contents, since biomass yield did not follow these same trends. In addition, there appears to be a close relationship between AMF inoculation, biomass yield and P accumulation, especially in locations with less P availability in the soil, as there was a higher P accumulation in the five areas tested when little P was available in the soil (Figure 4).

Grain yield is the most important variable for farmers, and yield increases may be a result of different benefits provided by the association with AMF (Rilling et al., 2002; Ryan and Graham, 2002; Ruiz-Lozano, 2003; Hart and Forsythe, 2012).

Higher biomass yield, stimulated or not by inoculant application, seems to be related to higher grain yield. In the present work, grain yield of soybean plants responded to phosphate fertilization (Figure 5). The highest grain yields were observed in the places with high natural P availability in the soil: 3.8 and 3.74 Mg ha^{-1} in RS and PR (Figure 5b and c), respectively, followed by MT and GO with 2.88 and 2.90 Mg ha^{-1} (Figure 5a and d), respectively, and lastly MG with 2.2 Mg ha^{-1} (Figure 5e), a site with very low natural P availability. However, in addition to responding to P doses, grain yield was stimulated by inoculation, and the average increase of inoculated plants over non-inoculated plants, in the five sites tested, was 29% (ranging from 15 to 46%) (Figure 5). This result proves the efficiency of the *R. intraradices* mycorrhizal inoculant for increasing grain yield for soybean.

These results corroborate other studies in which two (Meghvansi and Mahna, 2009) and four (Meghvansi et al., 2008) soybean cultivars were employed. The authors verified increases in seed weight when plants were inoculated with *R. intraradices*, proving once again the economic potential of using AMF-based inoculants for this crop. Considerations on other benefits provided by AMF in soybean were made by Porcel et al. (2003), who claim that inoculation with AMF mitigates, for example, the effect of water deficiencies on this crop.

Responses of AMF inoculation are dependent on crop type, area history and interrelationships between living communities and environmental factors. The results obtained here show that P-based controlled fertilization and the addition of *R. intraradices*-based inoculant resulted in a significant increase in grain yield for soybean. In addition to the direct effect on growth and yield, AMF may allow reductions in P rates applied to the soil, since it was observed that with 50% of the P dose, inoculated plants had the same grain yield as non-inoculated plants with 100% of the recommended P. Thus, this reduction has a direct effect on soil quality, reducing the dependence on phosphate fertilizers, as well as reducing the expenses related to the purchase of fertilizers by the farmer.

Conclusion

The *R. intraradices* mycorrhizal inoculant increased the biomass yield, P uptake and soybean yield under different edaphoclimatic conditions, with more marked effects on soils that originally had low or medium levels of available soil P. On average, inoculation provided an increase of 29% in biomass (regardless of the applied P dose) and grain yield, and higher P uptake. Grain yield of the inoculated plants was equal to or even greater than the non-inoculated plants that received 100% of the recommended P. This result suggests a better use of farmer's monetary resources, as well as indicates that the use of environmentally sound approaches, such as biological inoculation of plants, cannot only reduce farms inputs of fertilizers, but also contributes towards more sustainable farming.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Ardakani MR, Mazaheri D, Mafakheri S, Moghaddam A (2011). Absorption efficiency of N, P, K through triple inoculation of wheat (*Triticum aestivum* L.) by *Azospirillum brasilense, Streptomyces* sp., *Glomus intraradices* and manure application. Physiology and Molecular Biology of Plants 17:181-192.

Bago B, Pfeffer PE, Zipfel W, Lammers P, Shachar-Hill Y (2002). Tracking metabolism and imaging transport in arbuscular mycorrhizal fungi. Metabolism and transport in AM fungi. Plant and Soil 244:189-197.

Berruti A, Lumini E, Balestrini R, Bianciotto V (2016). Arbuscular Mycorrhizal Fungi as natural biofertilizers: let's benefit from past successes. Frontiers in Microbiology 6:1-13.

Bi YL, Wu FY, Wu YK (2005). Application of arbuscular mycorrhizal in ecological restoration of areas affected by coal mining in China. Acta Ecologica Sinica 25:2068-2073.

Bolan NS (1991). A critical review on the role of mycorrhizal fungi in the uptake of phosphorus by plants. Plant and Soil 134:189-207.

Brasil (2011). Ministério da agricultura, pecuária e abastecimento. Instrução normativa sda nº 13, de 24 de março de 2011. Altera a Instrução Normativa SARC no 05, de 6 de agosto de 2004. Diário Oficial da União: seção 1, sexta-feira, 25 de março de 2011.

Brasil (2013). Ministro de estado da agricultura, pecuária e abastecimento. Instrução normativa nº 53, de 23 de outubro de 2013. Que estabelece critérios de registro e produção de inoculantes. Diário Oficial da União: seção 1, 24 de Outubro de 2013.

Broch DL, Ranno SK (2012). Fertilidade do solo, Adubação e Nutrição da Cultura da Soja. In: Gitti e Roscoe Fundação MS, Tecnologia de Produção: Soja e Milho 2011/2012. Maracaju: Fundação MS. pp. 2-38.

Cely MVT, Oliveira AG, Freitas VF, Luca MB, Barazetti AR, Santos IMO, Gionco B Garcia GV, Prete CEC, Andrade G (2016). Inoculant of Arbuscular Mycorrhizal Fungi (*Rhizophagus clarus*) increase yield of soybean and cotton under field conditions. Frontiers in Microbiology 7:720.

Corrêa JC, Mauad M, Rosolem CA (2004). Fósforo no solo e desenvolvimento de soja influenciados pela adubação fosfatada e cobertura vegetal. Pesquisa Agropecuária Brasileira 39:1231-1237.

Crespo RJ (2015). Impact of arbuscular mycorrhizal fungi on the physiology of maize genotypes under variable nitrogen and phosphorus levels. Ph.D. Dissertation. University of Nebraska 150 p.

Dania SO, Fagbola O, Dania MI (2013). Response of Maize-Pigeon Pea intercrop to Arbuscular Mycorrhizal Fungi in nutrient depleted soil. Annual Review and Research in Biology 3:232-245.

Davison J, Moora M, Öpik M, Adholeya A, Ainsaar L, Bâ A, Burla S, Diedhiou AG, Hiiesalu I, Jairus T, Johnson NC, Kane A, Koorem K,. Kochar M, Ndiaye C, Pärtel M, Reier Ü, Saks Ü, Singh R, Vasar M, Zobel M (2015). Global assessment of arbuscular mycorrhizal fungus diversity reveals very low endemism. Science 349:970-973.

Diop TH (2003). In vitro culture of arbuscular mycorrhizal fungi: advances and future prospects. African Journal of Biotechnology 2:692-697.

Garg N, Chandel S (2010). Arbuscular mycorrhizal networks: process and functions, a review. Agronomy for Sustainable Development 30:581-599.

George E, Marschner H, Jakobsen I (1995). Role of Arbuscular Mycorrhizal Fungi in uptake of Phosphorus and Nitrogen from soil. Critical Reviews in Biotechnology 15:257-270.

Glomeromycota Species List (2019). [cited 2019 Nov 15]. Available from: http://www.amf-phylogeny.com/amphylo_species.html.

Guo W, Zhao R, Fu R, Bi N, Wang L, Zhao W, Guo J, Zhang J (2014). Contribution of arbuscular mycorrhizal fungi to the development of maize (*Zea mays* L.) grown in three types of coal mine spoils. Environmental Science Pollution Research 21:3592-3603.

Harley JL (1989). The significance of mycorrhizal. Mycological Research 92:129-139.

Harrison MJ (1995). Molecular and cellular aspects of the arbuscular mycorrhizal symbiosis. Annual Review of Plant Physiology and Plant Molecular Biology 50:361-389.

Hart MM, Forsythe JA (2012). Using arbuscular mycorrhizal fungi to improve the nutrient quality of crops; nutritional benefits in addition to phosphorus. Science and Horticulture 148:206-214.

Hodge A, Helgason T, Fitter AH (2010). Nutritional ecology of arbuscular mycorrhizal fungi. Fungal Ecology 3:267-273.

IBGE (2019). Levantamento Sistemático da Produção Agrícola. Tabela 1 - Área, Produção e Rendimento Médio - Confronto das Estimativas Setembro/Outubro – Brasil. [cited 2019 Nov 15]. Available from: https://www.ibge.gov.br/estatisticas/economicas/agricultura-e-pecuaria/9201-levantamento-sistematico-da-producao-agricola.html?=&t=resultados.

Jeffries P, Gianinazzi S, Perotto S, Turnau K, Barea JM (2003). The contribution of arbuscular mycorrhizal fungi in sustainable maintenance of plant health and soil fertility. Biology and Fertility of Soils 37:1-16.

Marschner H (1995). Functions of Mineral Nutrients: Macronutrients. pp. 229-314. In: Mineral nutrition of Higher Plants. Academic Press. 889 p. ISBN: 9780124735439.

Meghvansi MK, Mahna SK (2009). Evaluating the symbiotic potential of *Glomus intraradices* and *Bradyrhizobium japonicum* in Vertisol with two soybean cultivars. American-Eurasian Journal of Agronomy 2:21-25.

Meghvansi MK, Prasad K, Harwani D, Mahna SK (2008). Response of soybean cultivars toward inoculation with three arbuscular mycorrhizal fungi and *Bradyrhizobium japonicum* in the alluvial soil. European Journal of Soil Biology 44:316-323.

Oblinger JL, Koburger JA (1975). Understanding and Teaching the Most Probable Number Technique. Journal of Milk and Food Technology 38:540-545.

Ohtomo R, Saito M (2005). Polyphosphate dynamics in mycorrhizal roots during colonization of an arbuscular mycorrhizal fungus. New Phytologist 167:571-578.

Oliveira Jr A, Castro C, Pereira LR, Domingos CS (2016). Estádios fenológicos e marcha de absorção de nutrientes da soja. Embrapa Soja.

Orak AB, Demir S (2011). Effects of Arbuscular Mycorrhizal Fungus and Different Phosphorus Doses Against Cotton Wilt Caused by *Verticillium dahliae* Kleb. Plant Pathology Journal 10:108-114.

Porcel R, Barea JM, Ruiz-Lozano JM (2003). Antioxidant activities in mycorrhizal soybean plants under drought stress and their possible relationship to the process of nodule senescence. New Phytology 157:135-143.

Rezende PM, Carvalho ER, Santos JP, Andrade MJB, Alcantara HP (2009). Enxofre aplicado via foliar na cultura da soja [*Glycine max* (L.) Merrill]. Ciência e Agrotecnologia, 33:1255-1259.

Rillig MC (2004). Arbuscular mycorrhizae, glomalin, and soil aggregation. Canadian Journal of Soil Science 84:355-363.

Rilling MC, Wright SF, Eviner VT (2002). The role of arbuscular mycorrhizal fungi and glomalin in soil aggregation: comparing.

effects of five plant species. Plant and Soil 238:325-333.
Ruiz-Lozano JM (2003). Arbuscular mycorrhizal symbiosis and alleviation of osmotic stress. New perspectives for molecular studies. Mycorrhiza 13:309-317.
Ryan MH, Graham JH (2002). Is there a role for arbuscular mycorrhizal fungi in production agriculture? Plant and Soil Sciences 94:263-271.
Schuessler A (2015). System and methods for continuous propagation and mass production of arbuscular mycorrhizal fungi in liquid culture. Patent WO 2015028601 A1.
Sharif M, Ahmad E, Sarir MS, Muhammad D, Shafi M, Bakht J (2011). Response of different crops to Arbuscular Mycorrhiza Fungal inoculation in phosphorus-deficient soil. Communications in Soil Science and Plant Analysis 42:2299-2309.
Smith SE, Jakobsen I, Gronlund M, Smith FA (2011). Roles of arbuscular mycorrhizas in plant phosphorus nutrition: interactions between pathways of phosphorus uptake in arbuscular mycorrhizal roots have important implications for understanding and manipulating plant phosphorus acquisition. Plant Physiology 156:1050-1057.
Smith SE, Read DJ (2008). Mycorrhizal Symbiosis, 3rd Edn. London: Academic. 2008.
Smith SE, Smith FA (2011). Roles of Arbuscular Mycorrhizas in Plant Nutrition and Growth New Paradigms from Cellular to Ecosystem Scales. Annual Review of Plant Biology 62:227-250.
Sociedade Brasileira de Ciência do Solo (SBCS) (2004). Comissão de Química e Fertilidade do Solo – RS/SC. Manual de adubação e de calagem para os estados do Rio Grande do Sul e de Santa Catarina. 10a. ed. Porto Alegre.
Spagnoletti F, Lavado RS (2015). The Arbuscular Mycorrhiza *Rhizophagus intraradices* Reduces the Negative Effects of Arsenic on Soybean Plants. Agronomy 5:188-199.
Srinivasan M, Kumar K, Kumutha K, Marimuthu P (2014). Establishing monoxenic culture of arbuscular mycorrhizal fungus *Glomus intraradices* through root organ culture. Journal of Applied and Natural Science 6:290-293.
Tajini F, Trabelsi M, Drevon JJ (2012). Combined inoculation with *Glomus intraradices* and *Rhizobium tropici* CIAT899 increases phosphorus use efficiency for symbiotic nitrogen fixation in common bean (*Phaseolus vulgaris* L.). Saudi Journal of Biological Sciences 19:157-163.
Tedesco MJ, Gianello C, Bissani CA (1995). Análise de solos, plantas e outros materiais. Boletim Técnico de Solos. 5. 2. ed. Porto Alegre: UFRGS, 174 p.
Vosátka M, Látr A, Gianinazzi S, Albrechtová J (2012). Development of arbuscular mycorrhizal biotechnology and industry: current achievements and bottlenecks. Symbiosis 58:29-37.

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Full Length Research Paper

Effects of variable light intensities on seedling photomorphorgenesis and field performances of tomato (*Solanum lycopersicon* ‘Lindo F1)

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Six tomato (*Solanum lycopersicon* ‘Lindo F1) nursery shades and no shade control were evaluated for their effects on seedling photomorphogenesis and field performances as influenced by variable light transmission through the shades. The shades were constructed with wooden frames and different coloured polyethylene sheets as cover (roof). Different polyethylene colours showed varying effects on all the parameters measured. The control treatment (no shade) and green colour shade transmitted the highest light intensity while black and blue coloured shades received the highest temperature and no shade, the lowest. Early emergence and highest percentage emergence (68.8%) was observed under green shade colour compared to blue, red and non-shaded seedlings that gave 54.2, 60.4 and 52.5%, respectively. The environmental variables, light and temperature created microclimates in each of the shades that differed significantly and influenced physiological activities of the seedlings as evidenced in their yield parameters. Regression analysis revealed that light was more important than temperature in tomato plant growth and development. Evidence from the study revealed that the different coloured shades transmitted different light wave lengths to the seedlings in the nursery and consequently affected seedling performance in the field. Conclusively, tomato nursery under no shade treatment produced high quality seedlings that performed better under field conditions than those seedlings raised in the shade. If shades must be used, green and red colours which are capable of transmitting enough light waves for effective photosynthesis of the seedlings are recommended.

Key words: Light intensity, seedling, photomorphorgenesis, nursery, shade, photosynthesis.

INTRODUCTION

Tomato (*Solanum lycopersicon* L.) is one of the most widely grown vegetables in the world, an important

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condiment in most diets and a very cheap source of vitamins (Olaniyi et al., 2010). Tomato was introduced into West Africa by expatriate missionaries and civil servants (Uguru, 2011). It is a short-lived herbaceous plant with weakly woody stem that grows erect at the early stage but later slump to the ground due to the fruit weight. Usually, it grows 1 to 3 m or more above ground with well-developed taproots that branch profusely to form dense root systems (Uguru, 2011). Tomato plants are dicots, and grow as a series of branching stems, with terminal bud at the tip that does the actual growing. The vines are covered with fine short hairs that form or develop into roots whenever the plant makes contact with the ground and moisture.

Tomato products are generally stocked with vitamins (particularly vitamin C), minerals, fibers, antioxidants and phytonutrients that work together to help us stay healthy and fit. They are also a rich source of vitamin A and potassium (Jacy and Marilyn, 2003). Among their arsenal of nutrients, scientists are particularly intrigued with carotenoids, which are antioxidants that inactivate free radicals, slow progression of atherosclerosis, and protect us against cancer. Lycopene is what gives tomatoes their rich red colour. It is also what gives red colour to watermelon and pink grapefruit, but tomato and tomato products are the richest source of lycopene (Jacy and Marilyn, 2003).

Plants are capable of detecting and interpreting variety of environmental signals that influence their growth and development. Unique among these environmental signals is light. Sunlight satisfies two major important needs of biological organisms; energy and information. Radiant energy in the form of light from the sun is used by plant in manufacturing food through photosynthesis and to regulate movement, trigger developmental events and mark passage of time. Thus light is referred to as a narrow band of energy, with continuous electromagnetic spectrum emitted from the sun. In other words, it is the range of wavelength between 400 and approximately 700 nm. This range is defined as the photosynthetically active radiation (PAR), even though plants respond to other ranges of light like UV radiation (Hopkins and Huner, 2009).

The quantity and quality of light are constantly changing often in predictable ways. According to Hopkins and Huner (2009), plants are able to monitor these changes and make use of this information to direct growth and reproduction and the regulation of plant development by light is termed photomorphogenesis. The researchers further reported that plants have highly developed photo sensory systems comprised of light-sensitive photoreceptors and signal transduction pathways capable of acquiring and interpreting the information that is provided by light. A photoreceptor “reads” signal carried in the light by selectively absorbing different wavelength of light. This induces a conformational change in the pigment or an associated protein, photochemical oxidation reduction reaction, or some other form of photochemical change (Hopkins and Hunner, 2009). Whichever form the change takes, absorption of light normally induces a photochemical change that sets into motion a flow of events that ultimately results in developmental response. Therefore, manipulating any colour of the visible spectrum will influence plant developmental process.

Tomato yield is not an isolated characteristic and depends on the growth of the whole plant. If the tomato plant does not grow well after germination, then it will never give a high yield. Therefore yield is determined by the interaction between plant morphology, physiology and growth conditions (Van Der Ploeg and Heuvelink, 2005) It has been reported that different polyethylene colours used as shade reflect different spectra of the visible light and transmit some spectra of the visible light with consequent effects on physiological behaviors of plants. According to the author, varied light intensity causes varied light regulated developmental responses in plant which also influences the crop yield. Diverse light intensities applied during nursery would have an influence on different aspects of tomato growth and development, for example leaf and truss appearance, fruit growth period among other stages of developments (Van Der Ploeg and Heuvelink, 2005). Traditionally, tomato farmers adopt tree shades or constructed shades with plant materials for their nursery practices. The seedlings get indirect light transmission thereby forcing the seedlings to tilt towards the direction of sun light. Under this condition, most seedlings have their tips bent to more or less 45° during transplanting.

In the present work, we investigated the effects of diverse light intensities on growth, development and ultimately yield of tomato. We hoped to develop a nursery shade technique that farmers can employ with low demand on cost, skill and facilities to produce high yield. The specific objectives were to develop six micro climates using coloured polyethelene sheets as shade for tomato seedlings and to determine the effects of the micro climates on seedling growth in the nursery and final field growth and yield performance of tomato.

MATERIALS AND METHODS

Two experiments comprising nursery and field studies were conducted at the Department of Crop Science, University of Nigeria, Nsukka Research Farm between May and September 2017. Nsukka is located on latitude 06° 51’E and longitude 07° 29’N with an altitude of 475 m above sea level. It is characterized by lowland humid condition with bimodal annual rainfall distribution that ranges from 1155 to 1955 mm, a mean annual temperature of 29 to 31°C and a relative humidity that ranges from 69 to 79% (Uguru, 2011) in derived savanna ecology.

Nursery shades construction

The six nursery shades used were coloured polyethylene sheets comprising red, green, black, colorless, yellow and blue. The colored polyethylene sheets were of the same size and thickness. The shade for each experimental unit was constructed with 5 x 5 cm wooden bars each measuring 1.2 m long. One end of each wood was sharpened for easy pinning into the ground for support. Eight (8) pieces were used in the construction of each unit. The ground was first softened by watering before the pegs were pinned into the ground. They were driven into the ground perpendicularly by using a mallet to hammer the unsharpened end of the wooden poles until only 1 m length was left above ground (this makes for strong support). Footpath of 1 m wide was created among treatments as well as a 3 m gap between the surrounding vegetation and treatments. Each wooden frame was covered with different coloured polyethylene sheets according to the design to form a nursery shade (unit). The covering was from the top of the wooden frame to the ground in the direction of South-West wind to reduce damage to the polyethylene. However, on the other side receiving mild wind, the covering was left hanging 5 cm from the ground to allow for ventilation and to discourage influx of pest. The polyethylene sheets were fastened to each leg of the wooden frame with a twine to reduce effect of wind force on the seedlings. The top of the shade was perforated and tilted slightly to avoid collection of rain water on the top of the shade. There was a control frame without shade.

Nursery preparation

The poultry manure was dry and well cured as it was from battery cage. It was composted before use to reduce excessive heat build-up in the nursery. The medium was a conventional method prepared in the ratio of 3: 2: 1 of top soil, river sand and poultry manure respectively. The materials were thoroughly mixed and loaded into a wooden tray measuring 45 x 45 x 5 cm. About ¾ of each tray was filled with the medium materials. Seed viability test gave 98% germination before the tomato seeds were planted in grooves made on the medium in the trays. The trays were then put under each constructed shade treatment. The tomato variety used was “Lindo F1” marketed by Technisem - ZAC Anjou Actiparc de Jumelles - 49160 Longué-Jumelles – France.

Experimental design

The first experiment comprised of six coloured polyethylene shades and one control (no shade) treatment replicated three times in a completely randomized design (CRD) fashion. It was nursery experiment where each shade formed an experimental unit. The second experiment was set out in the field to evaluate the performances of the seedlings from the nursery. The land was mapped out into blocks and plots. Each plot measured 1.8 m long and 1.4 m wide with 1 m wide pathway on each side of the plot. Plant spacing was 60 cm between rows and 40 cm within rows.

Parameters measured in the experiment

Temperature (°C) and light intensity (Wm^{-2}) inside the nursery shade were measured. The temperature was measured using thermometer Model G11510 got from the Plant Pathology Laboratory of the Department of Crop Science, University of Nigeria, Nsukka. Light intensity was measured with a KIMO Solarimeter Model SAM 20:22493 obtainedfrom Energy Research Center, University of Nigeria, Nsukka. The reading was taken at daily interval for a period of four weeks and then the average was found.

Agronomic parameters such as days to seedling emergence, plant height, number of leaves, stem girth, internode lengths were measured in the nursery. Days to seedling emergence was determined by counting days between planting and plant emergence, while plant height was measured using meter rule and tailors’ measuring tape. Height measurement was taken from the growing medium surface to the top of the growing point on the stem (not the top of the leaf), number of leaves by counting; stem girth was measured with Micrometer screw gauge manufactured by Moore & Wright, Sheffield, England. A member of the NEILL group at two and four weeks after planting.

Field establishment stage

Site preparation

This stage of the experiment was to evaluate the field performance and yield of tomato seedlings raised under different coloured nursery shades. Poultry manure was applied liberally to the plots before transplanting. The manure was incorporated into the soil and allowed to further decompose for 5 days for the heat to dissipate before transplanting.

Transplanting

The seedlings were transplanted after 5 weeks in the nursery. Before transplanting, watering was reduced to a 3-day interval and the polyethylene sheets were removed 5 days before transplanting. This measure was taken to harden the seedling and reduce shock and stress after transplanting, and to gradually acclimatize them to the conditions of the field. The seedlings were well watered before transplanting to soften the soil so that the roots would not get damaged in the process.

Data collection and analysis

Parameters measured in the field were plant height at flowering, days to flowering for each sampled plant, days to fruiting, number of trusses per sampled plant at 2 and 4 weeks after transplanting (WAT), number of fruits per truss, number of fruits per plant at 2 and 4 WAT number of branches per plant, number of diseased fruits at maturity, number of fruits harvested per sampled plant, fruit length and circumference of each harvested fruit, days to fruit ripening, days to first harvest, weight of fruits harvested for each sampled plant. Fruit weight was measured using Furi Electronic Scale (Model No 7.1529) from Soil Science Laboratory, University of Nigeria.

All data collected were subjected to analysis of variance (ANOVA) following the procedures for CRD and RCBD using Genstat 4.0 Release 4.10.3DE. Treatment means were separated with F-LSD at 5% probability level. Graphical presentations were prepared with the Microsoft Excel chart wizard. Multiple linear regression analysis was done to determine the relationship between the environmental variables and yield parameters of the test crop. The backward elimination method was employed and the lower contributing variable was removed from the model in each case.

RESULTS

The amount of temperature and light intensity transmitted through the nursery shades varied (Figures 1 and 2). Generally, temperature under the polyethylene shades was higher than normal outside temperature especially during day time (Figure 1). The green polyethylene

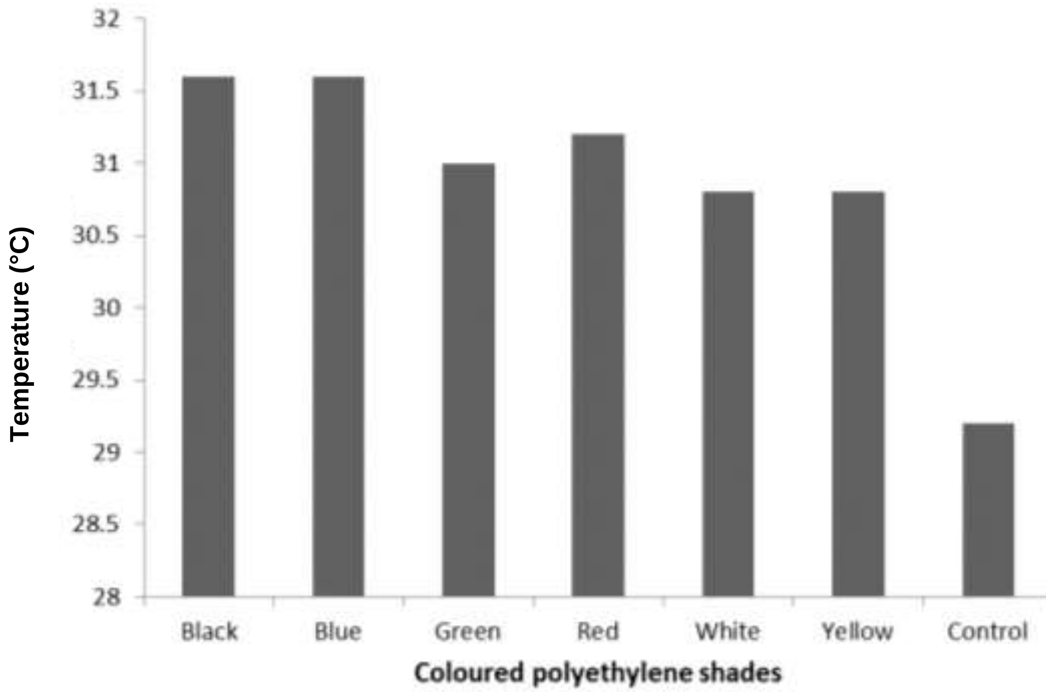

Figure 1. Average daily temperature (°C) reading inside the shades for five weeks.

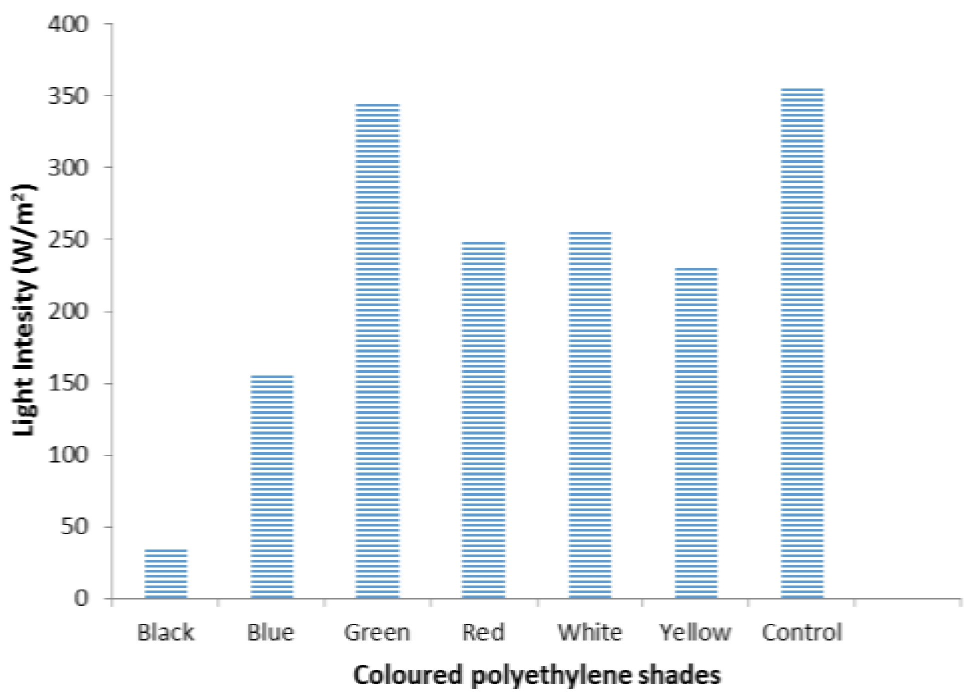

Figure 2. Average weekly light intensity (w/m^2) inside the shades for 4 weeks.

transmitted the highest light intensity, while black polyethylene transmitted by far the least light intensity of 35 w/m^2 followed by blue shade (157.5 w/m^2). The control treatment (no polyethylene shade) and green colour shade received the highest light intensity while black and blue coloured shades received the highest temperature

Table 1. Effect of nursery shade colour on seedling emergence pattern of tomato 'Lindo F1'.

Shade colour	Days to 1st emergence	Days to 50% emergence	Percentage emergence at 1st emergence	Percentage emergence at 1 week after planting	Percentage emergence at 2 weeks after planting
Black	5.67	-	9.24	-	-
Blue	5.67	9.00	12.50	25.00	54.20
Control	6.00	10.00	10.50	39.60	52.50
Green	5.67	7.00	27.10	53.80	68.80
Red	5.67	9.33	14.60	27.10	60.40
White	6.00	9.33	8.30	20.80	54.20
Yellow	5.67	8.00	29.30	41.70	66.70
LSD (0.05)	NS	1.81	17.48	27.67	21.74

Table 2. Effect of nursery shade colour on growth parameters of tomato 'Lindo F1' at 2 and 4 weeks after planting.

Shade Colour	Plant height at 2 weeks	Plant height at 4 weeks	Leaf count at 2 weeks	Leaf count at 4 weeks	Stem girth at 2 weeks	Stem girth at 4 weeks	Inter node length at 4 weeks
Black	-	-	-	-	-	-	-
Blue	6.42	12.82	8.44	21.60	2.40	4.56	3.52
Control	7.73	14.84	19.78	59.10	4.98	6.93	2.49
Green	6.99	14.50	9.44	23.30	3.17	5.15	3.97
Red	5.73	10.63	7.56	17.00	2.55	4.69	3.04
White	5.77	12.53	8.22	21.60	3.54	5.70	3.31
Yellow	7.71	16.82	11.20	30.80	3.38	5.60	4.19
LSD(0.05)	1.76	3.48	3.37	11.65	0.92	0.94	1.05

and no shade, the lowest.

The use of different polyethylene colours as nursery shade had no significant effects on seedling emergence six days after planting (Table 1). However, there was a significant ($P<0.05$) difference on the number of days to fifty per cent seedling emergence. The least number of days to fifty percent germination was recorded for seedlings grown under the green shade as it took only seven days to attain fifty percent germination. The yellow shade followed closely with less number of days, while blue, red and white colour shade seedlings showed about the same number of days to seedling emergence. The highest number of days to fifty percent emergence was recorded from the control while seeds planted under the black shade never attained fifty per cent emergence.

Seedling height, number of leaves and stem girth were significantly ($P<0.05$) affected by different polyethylene colours used as nursery shade at two weeks after planting. Seedlings grown under yellow shade and the non-shaded seedlings were the tallest compared to seedlings grown under red and white shades (Table 2). Seedlings grown under non-shaded frames had significantly ($P<0.05$) the highest number of photosynthetically active leaves of about (19.78) at this stage followed by yellow shade, while those grown under blue, green, red and white shades had the smallest which did not differ significantly. Tomato seedlings grown under different nursery shades responded differently to polyethylene colour effects at four weeks after planting. Seedlings grown under the yellow shade rapidly outgrew others and attained the tallest height and largest inter node length at four weeks after planting compared with blue colour and others (Plate 1a and b). There was no parameter measured for seedlings grown under the black polyethylene shade as the seedlings changed to a pale colour with spindle growth and died off at six days after germination.

The data regarding the field agronomic parameters of tomato plant are presented in Table 3. The seedlings raised in the green nursery shade produced the tallest plants (71.6 cm) compared to blue and un-shaded (no shade treatment). Plants from red, white and yellow shades were statistically similar to each other having heights of 68.4, 69.3 and 69.6 cm respectively. Plants from the control (unshaded) nursery flowered in a shorter period of 32 days followed closely by plants from green and white shades. Seedlings from unshaded (full sun) took the least number of days to fruit and for the fruit to get ripened compared to plants from blue shade nursery. Plants grown under the green shade significantly ($p \leq 0.05$) greater number of fruits at two weeks after transplanting while the plant grown under the blue shade consistently had the least number of flowers and fruits at two WAT. There was no significant difference in number

(a) (b)

Plate 1. Tomato seedling under a) yellow shade and b) blue shade.

Table 3. Effect of coloured polyethylene shade on field agronomic yield parameters of tomato 'Lindo F1'.

Shade colour	PHF (cm)	DFL	DFrt	DFrtRip	NF2wkT	NF4wkT	NT2wk	NT4wk	NBM
Black	-	-	-	-	-	-	-	-	-
Blue	63.40	46.50	57.50	88.44	11.11	23.22	2.778	6.89	3.89
Control	46.20	32.00	41.50	71.00	9.44	18.56	3.333	8.56	4.00
Green	71.60	34.40	39.20	84.78	13.22	24.22	2.778	7.22	3.56
Red	68.40	46.20	52.20	86.00	12.17	22.17	2.667	6.33	3.33
White	69.30	38.70	51.20	83.94	11.11	23.33	3.333	8.33	4.56
Yellow	69.60	46.00	57.20	87.17	12.72	29.33	2.778	8.11	4.50
LSD(0.05)	13.34	11.90	12.74	7.70	3.04	6.66	0.66	NS	NS

PHF= Plant height at flowering, DFL= Days to flowering, DFrt= Days to fruiting, DFrtRip= Days to fruit ripening, NF2wkT= Number of fruits at two weeks after transplanting, NF4wkT= Number of fruits at four weeks after transplanting, NT2wk= Number of trusses at two weeks after transplanting, NT4wk= Number of trusses at 4 weeks after transplanting, NBM= Number of ranches at maturity.

Table 4. Effect of nursery coloured polyethylene shade on yield components of Tomato 'Lindo F1'.

Shade colour	NDF4wk	NHF	NFL2wkT	FL(cm)	FC (cm)	TFW (g)	AvFW (g)	Yield (t/ha)
Black	-	-	-	-	-	-	-	-
Blue	4.39	4.17	9.90	5.006	14.88	252.915	61.00	3.50
Control	2.78	4.83	10.00	5.787	15.54	247.633	48.90	3.43
Green	3.00	6.00	21.70	5.965	17.24	499.348	90.20	6.94
Red	2.83	6.50	17.20	5.816	16.80	581.153	89.80	8.07
White	2.89	4.39	17.20	5.531	16.10	325.610	72.40	4.51
Yellow	3.44	6.72	16.30	5.223	15.74	452.608	71.30	6.27
LSD(0.05)	NS	NS	7.90	0.629	1.49	247.200	17.79	3.43

NDF4wk=Number of diseased fruit at 4 weeks after transplanting, NHF= Number of harvested fruit at maturity, FL=Fruit length, FC= Fruit circumference, TFW= Total fruit weight, AvFW= Average fruit weight, Yield= Yield in tons per hectare.

of fruits at two and four weeks after transplanting among the blue, white and green shaded plants. Number of trusses per plant was highest for plants grown under unshaded and white shaded frames after two weeks of transplanting compared to plants grown under blue, green, and yellow shades.

There was no significant effect on number of diseased fruits at four weeks after transplanting (Table 4). However, seedlings under control (no shade treatment) recorded the least disease incidence on the fruits in the field. Those seedlings under green shades produced fruits with the highest length and fruit circumference

Table 5. Model summary tables of multiple linear regression of environmental variables and tomato yield parameters.

Model	R	R Square	Adjusted R square	Standard error of the estimate
(a) Total fruit yield (t/ha)				
1	0.709[a]	0.503	0.255	1.517
2	0.673[b]	0.453	0.343	1.424
(b) Average fruit circumference				
1	0.732[a]	0.536	0.305	12.061
2	0.696[b]	0.485	0.382	11.371
(c) Average fruit length				
1	0.805[a]	0.647	0.471	0.249
2	0.673[b]	0.453	0.344	0.277
(d) Number of harvestable fruits				
1	0.589[a]	0.347	0.020	1.003
2	0.564[b]	0.318	0.182	0.917
3	0.000[c]	0.000	0.000	1.013

[a]Predictors: (Constant), Temp, light; [b]Predictors: (Constant), light; [c]Predictor: (constant).

(5.965 cm) and (17.24 cm) respectively compared to plants grown in blue polyethylene shade. Total fruit weight, average fruit weight and yield (t/ha) were significantly ($p \leq 0.05$) affected by different polyethylene colours shades used as nursery.

Multiple linear regression models with only 2 explanatory variables; light and temperature were entered in the model but temperature was always removed as a lower contributing variable as shown in Table 5a-c. Both light and temperature were removed from the model for number of harvestable fruits with almost the same values of R and R-Squared (Table 5d).

DISCUSSION

Factors other than light and temperature could influence seedling emergence and growth, but for this study only light and temperature were monitored. The study showed significant polyethylene colour effects on light transition, seedlings emergence, growth and development inside the nursery under the shades. Although, rate of photosynthesis was not measured in this study, seedlings in the no shade and green coloured shades that had the highest light transmission performed better in the field in terms of total yield suggesting that those seedlings had more photosynthetic activities than others. The highest and significant ($P \leq 0.05$) number of leaves produced by the no shade seedlings in this study can explain reasons for higher photosynthesis by seedlings in no shade treatment. It is also interesting here to note that active photosynthesis in the nursery had residual effects in the field performance due to good quality seedlings as influenced by different light intensity transmitted by the polyethylene colour in the nursery shades.

Variation in plant response to different light intensities is brought about by light attributes such as its intensity and wavelength (Baiyeri, 2006). It has been observed that the micro environment in which seedlings are grown has a major effect on growth and development of crops. This agrees with the findings of Hopkins and Huner (2009) which stated that the germination of seeds and survival of the seedlings that emerge are largely dictated by conditions in their immediate environment. Growth rate of crops at any instant time is related to earlier environmental effects on the plant biomass (Baiyeri 2006). Another study by Abdalbagi et al. (2010) disclosed that in the tropics, tomato is mostly produced through nursery and that good quality seedling usually leads to higher yield and early maturity in the field. It was also noted that poor quality seedling might not be improved by any means of management after being transplanted in the field. Thus seedlings that performed best in the nursery would likely grow better and faster when re-established in the field.

Observation from the study showed that emergence was poor for the black polyethylene shade, and the few plants that germinated showed a spindle growth with only two leaves which could not sustain and they finally slumped and died off after six days. This could be related to the insufficient light transmitted by the black polyethylene shade and inability of the plants to photosynthesize coupled with high temperature in the shade that made plant survival difficult. The best field performance of the plants from the green polyethelene shade could be attributed to the combination of light intensity and temperature under the green polyethylene which appeared to be the optimum for seedling development compared to other polyethylene covers. This agrees with the findings of Baiyeri (2006) in his study on pawpaw (*Carica papaya*) where he reported that the green polyethylene shade enhanced emergence and seedling quality in the nursery. Hopkins and Huner (2009) had earlier noted that absorption of light for normal photosynthesis induced a photochemical change that set into motion a flow of events that ultimately resulted in plant response to manipulation of any colour of the visible spectrum that influences plant developmental process. With respect to seedling height, leaf number and stem girth, seedlings grown under the no shade were superior in leaf number and stem girth at two and four weeks after planting while seedlings in the shade grew taller. This could be attributed to the inadequate light intensity in the shade while those outside shade received enough light intensity earlier for photosynthesis. These observations are supported by Thangam and Thangburaj (2008) who noted that plants grown under shade exhibited better growth in terms of plant height as compared to those in open field. However, the authors did not explain how they came about the result but light could be implicated.

Variation in fruit length, fruit circumference and average fruit weight of fruits from plants raised in different polyethylene sheets was observed in this study. The outstanding performances of plants raised in the green coloured nursery shade suggest that green shade possess the best microclimate for development of tomato seedlings. Tuzel et al. (2009) had earlier noted that environmental modification is important for tomato growth and development in the derived savannah agro-ecology where shading increased total yield per plant. Seedlings raised under the blue polyethylene shade had the highest number of diseased plants at four weeks after transplanting. This could be attributed to poor development of the seedlings as a result of low light intensity for adequate photosynthesis thereby making the plants weak and susceptible to disease attack (Hanson et al., 2000).

The result of regression analysis in this study showed that light is more important than temperature in tomato plant growth and development. This result corroborates Hopkins and Hunner (2009) assertion that plant utilizes light directly in biomass production through photosynthesis for all aspect of growth. It is evident in this study that tomato seedlings in the green polyethylene shades and control (no shade) that received up to 350 w/m^2 light intensities were highest in the yield parameters. The environmental parameters; light and temperature did not affect number of harvestable fruits. This is not surprising since only one variety of tomato with a particular genetic make-up was used. The result therefore, suggests that number of fruits produced by plants could be more of gene attribute than environmental effects.

Conclusion

It is evident in this study that difference in seedling morphogenesis and physiological quality of seedlings produced under the nursery shades evaluated were mainly due to variation in light intensity and temperature regimes under the shades. The higher yield parameters obtained in no treatment (no shade) nursery and green coloured shade could be accounted by the amount of light transmitted which probably resulted in greater photosynthesis of seedlings in these nurseries. It is also clear that the seedlings in the other shaded nurseries carried residual effects of low vigour to the field due to low accumulation of energy in the nursery that resulted from inadequate light transmission compared to green shade and open field. Conclusively, tomato nursery under no shade treatment gives better quality seedlings that withstand field conditions than those seedlings raised under shade. If shades must be used, green or red colour which are capable of transmitting enough light wave for effective photosynthesis to produce high quality seedlings is recommended based on our findings in this study.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Abdalbagi AH, Salih OS, Abdu EA, Ahmed M E (2010). Effect of Sowing Date and Plant Density on Growth and Yield of Tomato (Lycopersicon Esculentum, Mill.). Research Journal of Agriculture and Biological Sciences 6(5):665-669.

Baiyeri KP (2006). Seedling emergence and growth of pawpaw (*Carica papaya*) grown under different coloured shade polyethylene. International Agrophysics, pp. 77-84.

Hanson P, Chen J, Kuo C, Morris R, Opena R (2000). International Cooperator's Guide-Suggested Cultural Practices for Tomato. Shanhua: AVRDC-Asian Vegetable Research and Development Center. Retrieved from http://www.avrdc.org.tw

Hopkins WG, Huner NP (2009). Introductaion to plant physiology. John Wiley & Sons, Inc.

Jacy W, Marilyn OW (2003). Nutrition For Older Adults' Health. Athens: Department of Food and Nutrition, The University of Georgia.

Olaniyi J, Akanbi W, Adejumo T, Akande O (2010). Growth, fruit yield and nutritional quality of tomato varieties. African Journal of Food Science 4(6):398-402.

Thangam M, Thamburaj S (2008). Comparative perfromance of tomato varities and hybrids under shade and open conditions. Indian Journal of Horticulture 4:65.

Tuzel Y, Oztekin G, Meric M (2009). Influence of shading screens on microclimate, growth and productivity of tomato. Acta Horticulturae, pp. 97-102.

Uguru MI (2011). Crop Production: Tools, Techniques and Practice. Enugu: Fulladu Publishing Company.

Van Der Ploeg A, Heuvelink E (2005). Influence of sub-optimal temperature on tomato growth and yield: a review. Journal of Horticultural Science and Biotechnology, pp. 652-659.

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Determinants of sorghum productivity among small-scale farmers in Siaya County, Kenya

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The productivity of sorghum in Kenya is on decline despite sorghum being one of the suitable crops for the arid and semi-arid areas commonly found in Kenya. The study therefore aimed at establishing the effect of the selected socio-economic factors on sorghum productivity using a case of small-scale farmers in Siaya County. The four sub-counties considered for this study were selected on the basis of sorghum production. Stratified and random sampling techniques were applied to identify a sample comprising of 300 smallholder households in the study area. Data was collected using semi-structured interview schedules administered to the selected farm households. The characteristics of the smallholder farmers sampled were analyzed using descriptive statistics and Ordinary Least Square multiple regression model. The results showed that farm size under sorghum, labour, farm gate price, serena and seredo seed varieties were significant determinants of sorghum productivity in the study area. Based on these findings, the study recommends provision of improved seed varieties to the farmers. Policies targeted at promoting industrial use of sorghum will increase sorghum demand and promote its uptake. In addition, agricultural development policies should target provision of such services like training and extension support to enhance sorghum production in Kenya.

Key words: Sorghum productivity, arid and semi-arid, ordinary least square.

INTRODUCTION

Sorghum is one of the major cereal crops globally and is ranked fifth after maize, wheat, rice and barley in terms of production and importance (Naik et al., 2016). It is mainly grown in Central America, Africa and Asia primarily to enhance food security (Hassan, 2015). Globally, sorghum serves as a stable food for not less than 500 million people living in arid and semi-arid lands of Asia and Africa (Teferi, 2013). On average, sorghum constitutes 20% of the total cereals produced in Africa (Dube et al., 2014). Africa produces on average 20 million metric tons of sorghum which is a third of the global production of 60 million metric tons (Dube et al., 2014). In Eastern and Central Africa, sorghum cultivation covers an area close to 10 million ha. Nigeria is the leading sorghum producer in Africa at 34% seconded by Sudan at 21% (Mitaru et al., 2012). Other countries like Ethiopia, Tanzania, Uganda, Rwanda and Kenya account for 7, 4, 2, 0.8 and 0.6% respectively of sorghum produced in Africa (Mitaru

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et al., 2012).

Approximately, 43% of total landmass in Sub-Saharan Africa is arid and semi-arid and this is expected to increase as a result of climate change (Hadebe et al., 2017). In Sub-Saharan Africa, the predominant source of livelihood is rain-fed agriculture, mainly practiced on small-scale by smallholder farmers (Mavhura et al., 2015). Farmers usually realize very scanty yields from this system of farming especially during extreme drought periods (Mabhaudhi et al., 2017). It has been estimated that between 70-85% of the poor Africans entirely depend on agriculture for livelihood (Byerlee et al., 2005; Ravallion et al., 2007). It has also been reported that rural livelihoods face major food insecurity menace, 83% of the population being in the extreme poverty trap (FAO, IFAD, and WFP, 2014). Thus concerted efforts towards increasing agricultural productivity are necessary in Sub-Saharan Africa. There is still high potential of increasing both agricultural productivity and land productivity sustainably in this region, however, the fundamental question lies on how the Sub-Saharan Africa will realize its production potential (Conceição et al., 2016). One of the major entry point towards increasing agricultural production is the knowledge on food production dynamics especially in arid and semi- arid areas where small – scale farming is the major farming system, and farmers are less adaptive to climate change (Chivenge et al., 2015; Mundia et al., 2019). Sorghum plays a crucial role in providing food security in the face of climate change in many developing countries (Ogeto et al., 2013; Mundia et al., 2019).

Globally, there has been a positive growth in per capita food production over decades except in Africa where there is a steady decline in food production. Yield gap is therefore a major concern given that the global crop and food demand is projected to rise by more than 50% in 2050 (Pradhan et al., 2015). Arguably, since rain-fed agriculture occupies more than 95% of agricultural land use in Sub-Saharan Africa and water insufficiency is a real challenge, crops such as sorghum becomes major alternative crops in these regions since they are drought tolerant, require relatively less water, have high varieties of germplasm, have higher nutritional value (Hadebe et al., 2017), resistant to pest and diseases and adaptive to harsh arid and semi-arid environment (Chibarabada et al., 2017) as well reducing micro-nutrient mal-nutrition among farming households (Muui et al., 2013; Govender et al., 2017). Despite its potential in driving Sub-Saharan Africa towards sustainably increasing its food reserves, sorghum is under-utilized and production is still low (Orr et al., 2016; Grovermann et al., 2018). In some cases, sorghum has been associated with poverty, that is, some farmers tend to perceive sorghum as food for the poor (Dicko et al., 2006). These social factors together with other socio-economic and climatic factors have compounding effect on reducing uptake of sorghum as well as its production (Grovermann et al., 2018). In addition, Mwadalu and Mwangi (2013) pointed out that small – scale farmers have limited access to various institutional infrastructures such markets perpetuated by inadequate extension services and commodity processing facilities which negatively impact farm performance. Similarly, Rezig et al. (2010) highlighted that smallholder farmers are under- capacitated and face myriad of challenges, both biophysical and socio-economic in nature. Such constraints include low credit access, less fertile soils, inaccessibility to improved seed varieties and unreliable rainfall among others. This explains why low input agricultural systems, such as those of sorghum among other underutilized cropping systems, become essentially important in Sub- Saharan Africa (Rezig et al., 2010).

In contrary, the existing policies, Non-Governmental Organization aids and research on food crops generally tend to give high priority to major crops such as maize and as such encouraging production of crops less adapted to arid and semi-arid areas (Mukarumbwa and Mushunje, 2010; Pingali, 2015). Enhancing crop diversification through active inclusion of underutilized and drought tolerant crops such as sorghum and millet in order to boost farmer resilience and adaptive capacity to climate change are important across Africa and Asia (Fischer et al., 2016). As a result, there is need for a paradigm shift with respect to government agricultural policy setting, research and funding, meaning that, focus should be on diversified agriculture rather than agricultural system that promote a few selected crops (Massawe et al., 2015; Fischer et al., 2016; Mabhaudhi et al., 2017). A change in policy should promote climate responsive agriculture that encourages production of drought tolerant crops in arid and semi-arid areas in developing countries (Fischer et al., 2016). In addition, realization of sustainable food production will require that the aim of the current research should go beyond improving production but also to matching crops with the suitable agro-ecology (Sebastian, 2009).

In Kenya, sorghum is mainly grown in Nyanza, Eastern and Coastal regions which are frequently affected by drought and crop failure. Sorghum being an indigenous crop in Kenya is more suitable in salvaging households from hunger and poverty (Muui et al., 2013). However, the production of sorghum is on decline and the yields obtained by farmers are at unsatisfactory levels of 0.85 tons per hectare (Muui et al., 2013). The acreage apportioned to sorghum is also low compared to other crops such as maize (Kilambaya and Witwer, 2013). Similarly, the production of sorghum and millet is below the consumption demand (Recha, 2018). Masese et al. (2018) indicated that there is sorghum consumption deficit of more than 60 metric tons. The yield gap stands at approximately 70 000 tons due to decline in national sorghum production from 189 000 tons in 2015 to 117 000 metric tons in 2016 (Food and Agricultural Organization Statistics (FAOSTAT), 2018).

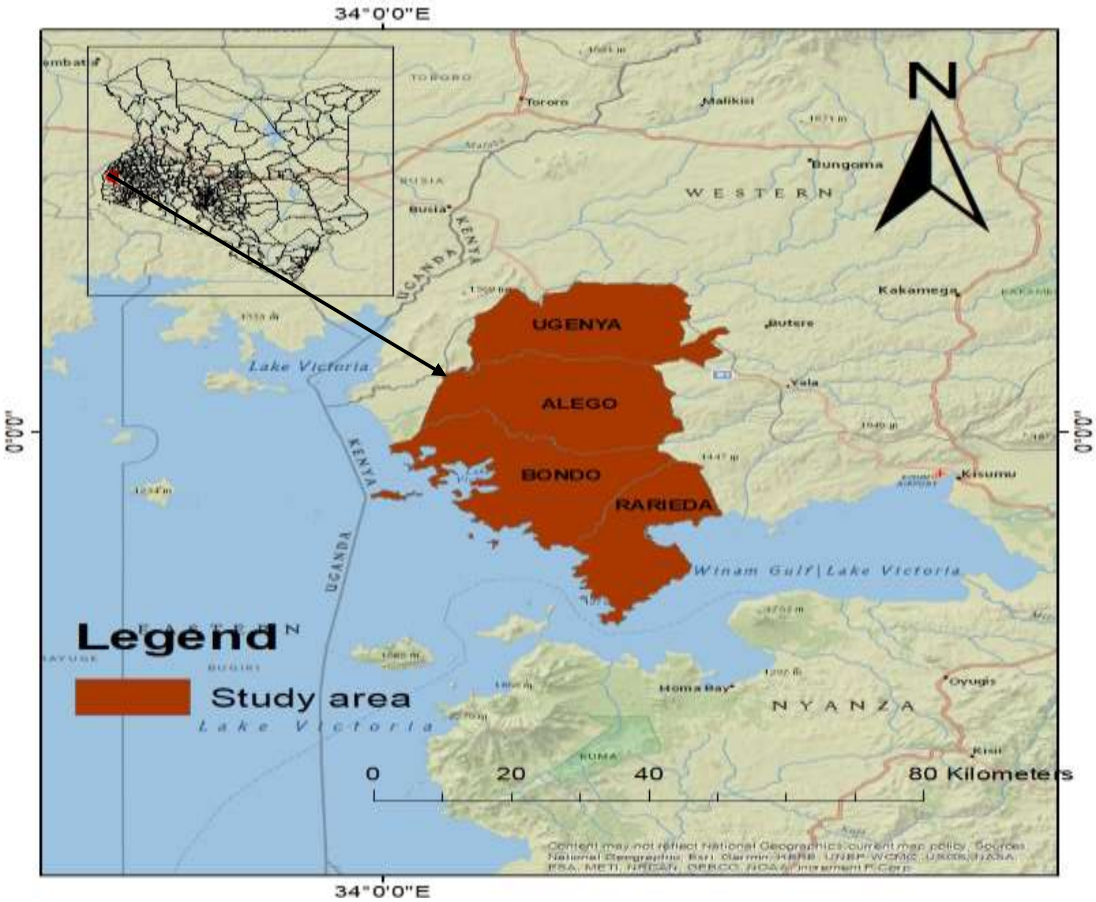

Figure 1. Map of the study area.

Several interventions have been made towards increasing sorghum production. Such interventions include release of drought, disease and pest tolerant High Yielding Sorghum Varieties (HYSVs) by research institutions like the International Crop Research Institute for Semi-Arid Tropics (ICRISAT). However, optimal sorghum production has not been achieved, implying increasing production does not necessarily rely on new technology uptake alone (Karanja et al., 2009). Production variations among farmers may be due to differences in regional settings and socio-economic characteristics of the farmer (Chimai, 2011). It is equally important to note that, despite their importance, there is inadequate research based information on underutilized crops such as sorghum (Mukarumbwa and Mushunje, 2010; Mabhaudhi et al., 2016; Orr et al., 2016; Mabhaudhi et al., 2017; Mundia et al., 2019) since most research have been focused on major cereal crops such as maize, rice and wheat. The study therefore aims to analyse the effect of the selected factors on the overall sorghum productivity unlike the previous studies that were specific to the effeciency of production (Chepng'etich et al., 2014, 2015; Zalkuwi et al., 2015; Naim et al., 2017; Okuyama et al., 2017; Botiabane et al., 2018).

METHODOLOGY

Description of the study area

The study was conducted in Siaya County. The county is located in Nyanza region, western Kenya. It borders Kakamega and Vihiga counties to the north-east, Busia County to the north, Kisumu County to the south-east and Lake Victoria to the south. The area lies between latitude 0°26' to 0°18' north and longitude 34°58' east and 33°33' west (Figure 1). The population of the county is approximately 841,682 people (GoK, 2009). The predominant agro-ecological zone (AEZ) in the county is lower midland zones (LM) starting from LM 1 up to LM 5 with traces of upper midland zones (UM) in areas with high agricultural potential (MoALF, 2016). The area is characterized by an annual mean temperature of 22.3°C with rainfall ranging from 900 to 1500 mm per year. Soil types range from low nutrient soil to red volcanic soil in some areas. Some of the major crops grown include maize, sorghum, beans, cowpeas, millet, sweet potatoes, groundnuts, cotton and sugarcane (MoALF, 2016).

Research design and sampling procedure

The study employed cross-sectional survey design. This research design enables the researcher to collect data at single point in time. The study applied a combination of two-stage stratified random sampling and probability proportionate to size sampling techniques in selecting the sample households. In the first stage, the four sub-counties were selected purposely on the basis of sorghum production. Once the sub-counties had been selected, a two-stage stratified sampling technique was applied to select one ward and subsequently one village from each ward in every sub-county selected. Finally, households were randomly selected from each of the randomly selected villages to achieve the target sample size. The number of households selected from each village was determined by the population of that village.

Data collection, sample size and analysis

Data for this study was collected using semi-structured interview schedules. A total of 300 farmers from the selected households were interviewed from the three sorghum producing sub-counties, namely; Rarieda, Bondo, Alego and Ugenya (Figure 1). Prior to data collection exercise, the data collection instruments were tested both for validity and reliability using a sample of 20 interview schedules administered to 20 randomly selected households. By applying split-half method, the instrument yielded a reliability coefficient of 0.7, thus proving reliable. The items found to be ambiguous and inadequate were correctly worded and re-modified to improve clarity. This study used primary data. The information collected was on household demographics such as gender, age, education, household size, total monthly income, off-farm income; farm characteristics such as yields, labour, farm experience and farm size and institutional factors such as extension contact, training, group membership, market access, road access, credit access and seed varieties. The data collected was analyzed using descriptive statistics (mean) and Ordinary Least Square (OLS) applied for descriptive and quantitative analysis respectively.

OLS model specification

Stochastic OLS multiple regression model was used to determine the effect of the selected factors on sorghum productivity. OLS is the most widely used economic model in estimating beta (β) parameters in several quantitative analysis (Rutherford, 2001). OLS multiple regression model shows the relationship between multiple independent variables and a continuous dependent variable. It is regarded as one of the most powerful statistical model in statistical analysis because of its relative simplicity in checking model assumptions such as linearity, constant, variance and the effect of outliers (Hutcheson and Sofroniou, 1999). The model shows the magnitude of the effect of the independent variable on the dependent variable. OLS model is linearly specified in the form;

$$Y_i = \beta_0 + \beta_i X_i \dots \beta_n X_n + U_i \quad (1)$$

Where Y_i is the dependent variable, β_0 is the intercept, $\beta_i \dots \beta_n$ are the coefficients of explanatory variables, $X_i \dots X_n$ are the explanatory variables while U_i is the error term. Based on Equation 1, we specified our model as;

$$Y_i = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \beta_4 X_4 + \beta_5 X_5 + \beta_6 X_6 + \beta_7 X_7 + \beta_8 X_8 + \beta_9 X_9 + \beta_{10} X_{10} + \beta_{11} X_{11} + \beta_{12} X_{12} + \beta_{13} X_{13} + \beta_{14} X_{14} + \beta_{15} X_{15} + \beta_{16} X_{16} + \beta_{17} X_{17} + \beta_{18} X_{18} + \beta_{19} X_{19} + \beta_{20} X_{20} + U_i \quad (2)$$

Where, Y_i represents sorghum quantity (dependent variable) X_1 represents age (years), X_2 represents gender of respondent (years), X_3 represents education of respondent (years), X_4 represents household size (number of household members), X_5 represents total monthly income (KES), X_6 represents monthly off-farm income (KES), X_7 represents land size (acres), X_8 represents labour (man days), X_9 represents hired labour (man-days) X_{10} represents family labour (man-days), X_{11} represents farm gate price (KES), X_{12} represents market distance (kilometres), X_{13} represents land ownership (dummy), X_{14} represents credit access (dummy), X_{15} represents training access (dummy), X_{16} represents seredo variety (dummy), X_{17} represents gadam variety (dummy), X_{18} represents serena variety (dummy), X_{19} represents ochuti variety (dummy), X_{20} represents nyakabar variety (dummy), β_0 to β_{19} are the parameters to be estimated and U_i is the error term, randomly distributed.

RESULTS AND DISCUSSION

Descriptive statistics

The selected variables for the study were summarized and presented in Table 1. On average, farmers produced 555.31 kg/acre which is approximately 0.555 tons/acre. This implies that the current sorghum production level in the area is far much below the potential output of 1.52 tons/acre (Muui et al., 2013) or 2-5 tons/acre (FAOSTAT, 2018). The mean age of the farmers was 47.13 which is comparable to the findings of several previous studies (Danso-Abbeam et al., 2018; Mmbando and Baiyegunhi, 2017; Suvedi et al., 2017; Ghimire et al., 2015); implying that farming is mostly dominated by the elderly. More male farmers (69%) than female farmer constituted the sample. The average years of schooling were 9 years with the majority of the sampled households constituting of 5 persons. Large family sizes are a likely implication that households have readily available farm labour derived from the family. Most of the households had a total monthly income of KES. 11, 336.68 and an average monthly off-farm income of KES. 8, 111.56. This shows that the largest share of the households' income come from off-farm income sources; implying that majority of the households depend on off-farm employment for livelihood alongside agriculture. Households allocated an average land size of 0.74 acres to sorghum. In regard to labour use, 65 man days on average was used in one acre of sorghum. The average farm gate price of sorghum per kilogram was KES. 32. In terms of proximity to the market, the average distance to the nearest market was 2 km. This implies that farmers incurred less transport cost when shipping the produce to the nearest market. In addition, since market and road distance can be used as proxy for input accessibility, nearness to the

Table 1. Descriptive results of variables and the expected effects on sorghum production.

Variable	Description	Mean	SD	Expected sign
Dependent variable				
Sorghum productivity	Quantity of sorghum in kilogram per acre	555.31	385.18	
Independent variable				
Age (years)	Age of the respondent in years	47.13	14.25	-
Gender	1, if the gender of the respondent is male; 0, if otherwise	0.69	0.48	+/-
Education (years)	Number of years spent in school by respondent	9.03	4.27	+
Household size	Total number of household members directly dependent on household head	5.40	2.80	+
Household income (KES)	Average total monthly income	11336.68	9057.48	+
Off-farm income (KES)	Average monthly off-farm income	8111.56	9278.24	+/-
Land size (acres)	Total farm size under sorghum	0.74	0.61	+/-
Labour (man-days)	Labour used per acre of sorghum	65.23	36.07	+/-
Farm gate price	Farm gate price of sorghum per kilogram	32.42	7.18	+/-
Market distance (km)	Distance to the nearest market	2.17	1.51	+/-
Land ownership	1, if the land under cultivation is owned by the household; 0, if rented	0.93	0.25	+/-
Training	1, if the household receives training; 0, if otherwise	0.41	0.49	+
Credit access	1, if the household has access to credit; 0, if otherwise	0.33	0.47	+/-
Seredo seed variety	1, if the household grows seredo variety; 0, if otherwise	0.45	0.50	+
Gadam seed variety	1, if the household grows gadam variety; 0, if otherwise	0.05	0.22	+
Serena seed variety	1, if the household grows serena variety; 0, if otherwise	0.05	0.21	+
Ochuti seed variety	1, if the household grows ochuti variety; 0, if otherwise	0.22	0.41	-
Nyakabar seed variety	1, if the household grows nyakabar variety; 0, if otherwise	0.05	0.23	-

Source: Own survey (2019); Note: 1 USD is an equivalent of KES. 100 at the time of the study; SD denotes standard deviation.

market increases farmers’ access to farm inputs. In terms of land ownership, 93% of the households operated on own lands; implying that most households are able to adopt better farming systems and technologies due to high tenure security and absence of land rental fee. Most of the households (59%) did not receive farm related training. In regard to credit access, 67% of the sampled farmers reported lack of access to farm credit. This explicitly details limited existence of institutional services meant to improve agricultural production in the area. Compared to other varieties, seredo variety was the most adopted as represented by 55% of the farmers. Serena, gadam and nyakabar were the least adopted. Only 0.05% of the farmers produced these varieties.

The mean sorghum yield was high among male than female farmers (Table 2). Generally, women are more disadvantaged in regard to farm credit accessibility (Adebayo and Adenkule, 2016). The output quantity is always lower for women than men due to limited access to extension services, credit and capital among female farmers (Ayelech, 2011). These disparities in terms of access to production services explain the significant variations in production across different gender. We found a significant difference in the quantity of sorghum produced between family and hired labour (Table 2). This implies that the mean sorghum output was high for hired labour than family labour.

Multiple regression results

The regression analysis was done using the STATA software and OLS multiple regression model. The results of the multiple regression are given in Table 3 and discussed hereafter.

Multicollinearity test

Multicollinearity exists when the explanatory variables are highly linearly correlated such that it is quite difficult to differentiate between which independent variables (X) affect dependent variable (Y) (Midi et al., 2010). Existence or non - existence of multicollinearity is explained based on the values of Variance Inflation Factor (VIF). The VIF value of the predictor variables should neither be greater than 10 nor less than one (Gujarati, 2003). VIF value greater than 10 indicates multicollinearity (Allison, 2001). It can therefore be concluded that multicollinearity exist if the VIF values exceeds 10 or smaller than 1. The results presented (Table 3) showed that there was no multicollinearity since none of the variable had VIF less than 1 or greater than 10.

Table 2. Productivity comparison between gender, age groups and labour source.

Variable	Productivity per acre	SD	t	Sig.
Gender				
Male	606.21	334.44	1.681	0.094**
Female	513.04	407.25		
Age categories				
18 - 35	496.46	362.08	-1.186	0.237
> 35	568.69	389.01		
Labour source				
Family	433.05	272.18	-4.697	0.000**
Hired	736.64	449.98		

Source: Own survey (2019); *** Sig. 1%, ** sig. 5% and *sig. 10%.

Table 3. Multiple regression results on selected factors affecting sorghum productivity (kilograms of sorghum per acre).

Sorghum quantity	Coef.	Std. Err.	t	P > |t|	VIF
Social factors					
Age	-0.013	0.051	-0.26	0.796	1.14
Gender	0.018	0.014	1.27	0.205	1.16
Education	0.004	0.006	0.59	0.553	1.33
Household size	0.024	0.033	0.73	0.469	1.40
Household income	-0.018	0.025	-0.73	0.467	1.90
Off-farm income	-0.008	0.010	-0.78	0.439	1.15
Economic factors					
Land size	-0.824	0.060	-13.67	0.000***	6.95
Labour (man days)	0.170	0.062	2.76	0.006***	3.95
Hired labour	0.035	0.018	1.94	0.053*	2.33
Family labour	-0.002	0.016	-0.15	0.880	2.34
Farm gate price	0.389	0.017	23.53	0.000***	3.40
Institutional factors					
Market distance	-0.005	0.165	-0.32	0.753	1.16
Land ownership	0.006	0.008	0.77	0.444	1.14
Credit access	0.006	0.014	0.43	0.671	1.94
Training	0.017	0.136	1.23	0.221	1.90
Seredo variety	-0.041	0.016	-2.54	0.012**	2.21
Gadam variety	-0.045	0.035	-1.26	0.208	1.36
Serena variety	-0.034	0.018	-1.88	0.062*	1.34
Ochuti variety	-0.003	0.018	-0.14	0.887	1.87
Nyakabar variety	0.021	0.026	0.82	0.411	1.38
R-squared = 0.94					

Source: Own analysis (2019); ***Sig. 1%, **sig. 5% and *sig. 10%.

Heteroscedasticity and endogeneity test

The heteroscedasticity and endogeneity tests were done to check whether the OLS multiple regression model assumptions were violated. This study employed Breusch-Pagan and Koenker test to check for the presence of heteroscedasticity. The Breusch-Pagan test value (LM = 22.207; p = 0.052) and Koenker test value (LM = 11.955; p = 0.531) showed that there was no existence of heteroscedasticity. This was achieved after subjecting the outcome variable and some predictor variables to logarithmic transformation.

Endogeneity exists when the explanatory variable is correlated with the error term resulting to inconsistency and biasness of the parameter estimates of the model. Endogeneity could result from either error related to measurement of a variable or variable omission (Maddala and Lahiri, 1992). The results of endogeneity using Durbin and Wu-Hausman test yielded Durbin score ($p = 0.7593$) and Wu-Hausman score ($p = 0.7733$). Therefore, the null hypothesis for exogeneity was accepted, implying that the quantities of sorghum produced were exogenous.

Selected factors affecting sorghum productivity

Table 3 shows the relationship between the study explanatory variables and sorghum productivity. The model accounted for 94% variations in sorghum production (Table 3). Land size, labour, source of labour (hired labour), farm gate price, seredo seed variety and serena seed variety were found to have significant contribution to sorghum productivity. On the other hand, age, gender, household size, total monthly income, monthly off-farm income, market distance, credit access, training and other selected seed varieties had insignificant effect on sorghum productivity.

Land size had significant effect on sorghum productivity ($p = 0.000$, $t = -13.67$) at 1% level of significance. We found that there was inverse relationship between sorghum productivity and land size, that is, productivity reduced by a factor of 0.824 (Table 3) per one acre increase in land size. This negative relationship between farm size and productivity confirms the results of Hazell and Hangbladde (2010) and Birachi et al. (2013) that smaller farms tend to be more productive than larger farms. The reciprocal relationship between farm size and productivity can be ascribed to the fact that small farms are easily manageable to poor resource-constrained small-scale farmers. Increase in farm size is associated with increased input requirement and input use which might be unsustainable to most of the small-scale farmers. Significant effect of land size on productivity has also been reported from other studies (Obasi and Ukewuihe, 2013; Ayoola et al., 2016; Abdulrahman et al., 2018; Daudi and Omotayo, 2018) in contradiction to the findings of (Anyaegbunam et al., 2012).

On the other hand, labour in general, that is, irrespective of the source, was found to be significant and positively related with sorghum productivity ($p = 0.006$, $t = 2.76$) at 5% level of significance. An increase in man-day was found to increase productivity by a factor of 0.170 (Table 3). This is consistent with the findings of Katundu et al. (2014); Dessale (2018) and Ombuki (2018). In regard to the specificities of labour source, hired labour positively contributed to sorghum productivity ($p = 0.053$, $t = 1.94$) at 10% level of significance. This shows that an additional labour from hired labour source increases sorghum productivity by a factor of 0.035. A study in Europe on the productivity of labour reported that farms that relied on hired labour were more productive than those that purely used family labour (Kloss and Petrik, 2018). Conversely, in Bangladesh, family labour was found to be more productive than hired labour (Chowdhury, 2016). In addition, a study conducted in Malawi reported no significant effect of family labour offered by children on productivity whereas family labour offered by male and female adults negatively affected productivity (Assefa, 2010). The same study however, reported a significant and positive effect of hired labour on productivity, reflecting the findings of the current study. On contrary, family labour was insignificant and had a negative relationship with sorghum productivity. The inconsistencies in family and hired labour effect on productivity could be attributed to the differences in the socio-economic characteristics and geographical settings of the farmers. Alternatively, the positive effect of hired labour on productivity could be attributed to higher efficiency of hired labour than family labour.

Among the seed varieties used, seredo and serena varieties significantly contributed to sorghum productivity. Contrary to the expectation, a negative relationship was observed between these varieties and sorghum productivity. A decrease in productivity by a factor of 0.041 and 0.034 was observed as a result of using seredo and serena varieties respectively. The negative relationship between these varieties and sorghum productivity could be attributed to use of overstocked seeds, untimely planting, inadequate soil fertilization due to financial constraints and yield loss to birds all of which have adverse effect on the productivity of both local and new varieties. Low farm productivity in Siaya County has also been attributed to limited access to and use of certified seeds and fertilizer in the previous study (Obiero, 2013). This is consistent with the findings of this study, where only a smaller proportion of the farmers had adopted the use of improved sorghum varieties (Table 1). Similarly, Urassa (2017) highlighted limited access to improved seed varieties and fertilizer as a major constraint towards improving productivity. The productivity of sorghum has been adversely affected by birds. For instance, 60% grain loss to birds has been reported in eastern Kenya (Orr et al., 2013; Hiron et al., 2014). In addition, despite the use of improved sorghum seed varieties, a study conducted in three sorghum producing regions, namely; Machakos, Kitui and Makueni in Kenya reported a significant grain loss to birds. Approximately, 80, 51 and 14% yield loss to birds was reported in Machakos, Makueni and Kitui respectively (Mutisya et al., 2016). As a coping mechanism, most farmers in arid areas have prioritized drought tolerant local varieties over some new varieties which are less resistant to drought and birds (Amelework et al., 2016).

Farm gate price was significant ($p = 0.000$, $t = 23.53$)

and positively related with sorghum productivity. We found that a unit increase in price was associated with 0.389 units increase in sorghum productivity. Output price influences farmers' adoption decision as well as resource allocation due to anticipated returns. Thus positive relationship between productivity and output price can be credited to increased resource allocation such as use of fertilizer which boosts sorghum's productivity. An interaction between the farm output price and the total factor productivity which interdependently affect farm profitability has been reported in the previous studies (Mugera et al., 2016).

Conclusion

The aim of this study was to analyze factors affecting sorghum productivity among small-scale farmers. Among the factors studied, land size under sorghum, labour, farm gate price and sorghum seed varieties used, that is, seredo and serena varieties were found to have significant effect on the overall sorghum productivity.

The study recommends that any policy intervention on sorghum production should prioritize the aforementioned significant factors. The study advises on the need for land redistribution as implicated by the inverse land size-productivity relationship. There is need for the county governments to provide machinery incentives such as tractors services at affordable hire charges to help during land preparation. This will reduce the cost of labour and the number of man hours required during land preparation. In order to achieve better prices for sorghum, the study emphasizes on the need to promote industrial use of sorghum, such as blending sorghum with other crops, for instance, sorghum-cassava floor. This will increase the industrial demand for sorghum and consequently better prices. This is likely to stimulate commercialization of sorghum production and make the crop competitive at the market. The seed varieties used, particularly serena and seredo varieties impacted on the overall productivity of sorghum. However, there was a negative relationship which could be accredited to limited use of improved sorghum seed varieties, over reliance on overstocked seeds extra. The study recommends increased support to farmers through regular provision of certified seeds and training to avert production constraints related to untimely planting, access to improved seed varieties and grain loss to birds.

Future research needs to lay focus on the marketing strategies adopted by the sorghum farmers and the associated transaction costs incurred across the various channels of marketing. This will help in designing policies targeted towards making sorghum enterprise more profitable. Further studies should also incorporate the effect of other factors such as spacing, fertilizer use, and pest and weed control on sorghum productivity at the farm level.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Abdulrahman S, Yusuf O, Sanni SA, Ayinde TB (2018). Technical Efficiency of Cocoyam Production in Kaduna State, Nigeria : A Stochastic Production Frontier Approach. Journal of Economics Management and Trade 21(11):1-10.

Adebayo SA, Adenkule OA (2016). Socio - economic status of women in group membership in selected areas of Kwara State, Nigeria. Agrosearch 1:59-67.

Amelework BA, Shimelis HA, Tongoona P, Mengistu F, Laing MD, Ayele DG (2016). Sorghum production systems and constraints, and coping strategies under drought-prone agro-ecologies of Ethiopia. South African Journal of Plant and Soil 33(3):207-221

Anyaegbunam HN, Nto PO, Okoye BC, Madu TU (2012). Analysis of Determinants of Farm Size Productivity among Small- Holder Cassava Farmers in South East Agro Ecological Zone, Nigeria. American Journal of Experimental Agriculture 2(1):74-80.

Assefa TW (2010). Raising labour productivity to solve the paradox; labour shortage in the labour surplus economy; Malawi. (Is targeted fertilizer subsidy the solution?) (Master's thesis).

Ayelech T (2011). Market Chain Analysis of Fruits for Gomma Woreda, Jimma Zone, Oromia National Regional State. MSc Thesis, Haramaya University, Haramaya, Ethiopia

Ayoola JB, Dangbegnon CK, Daudu A, Mando TM, Kudi IY, Amapu JO (2016). Socio-economic factors influencing rice production among male and female farmers in Northern Guinea Savanna Nogeriin action researcha: lessons for promoting gender equity. Agriculture and Biology Journal of North America, pp. 2151-7517.

Botiabane MP, Zhou L, Oluwatayo IB, Oyedokun FO, Oyelana AA (2018). Socioeconomic Analysis and Technical Efficiency among Smallholder Sorghum Farmers in GaMasemola Township of Limpopo Province, South Africa. Journal of Economics and Behavioral Studies 9(6):17.

Byerlee D, Diao X, Jackson C (2005). Agriculture, Rural Development, and Pro-poor Growth: Country Experiences in the Post-Reform Era Contribution of Agricultural Households to National Poverty Reduction in Africa. Agriculture 21:1-15.

Chepng'etich E, Nyamwaro SO, Bett EK, Kizito K (2015). Factors that influence technical efficiency of sorghum production: A case of smallholder sorghum producers in Lower Eastern Kenya. Advances in Agriculture, 2015.

Chepng'etich E, Bett EK, Nyamwaro SO, Kizito K (2014). Analysis of technical efficiency of sorghum production in lower Eastern Kenya: A Data Envelopment Analysis (DEA) approach. Journal of Economics and Sustainable Development 5(4):58-65.

Chibarabada TP, Modi AT, Mabhaudhi T (2017). Expounding the value of grain legumes in the semi- and arid tropics. Sustainability 9(1):60.

Chimai BC (2011). Determinants of technical efficiency in smallholder Sorghum farming in Zambia (Doctoral dissertation, The Ohio State University).

Chivenge P, Mabhaudhi T, Modi AT (2015). The Potential Role of Neglected and Underutilised Crop Species as Future Crops under Water Scarce Conditions in Sub-Saharan Africa. International Journal of Environmental Research and Public Health 12:5685-5711.

Chowdhury NT (2016). The relative efficiency of hired and family labour in Bangladesh agriculture. Journal of International Development 28(7):1075-1091.

Conceição P, Levine S, Lipton M, Warren-rodríguez A (2016). Toward a food secure future : Ensuring food security for sustainable human development in Sub-Saharan Africa. Journal of Food Policy 60:1-9.

Daudi AS, Omotayo AO (2018). Socioeconomic determinants of rural households' food crop production in Ogun state, Nigeria. Applied Ecology and Environmental Research 16(3):3627-3635.

Dessale M (2018). Measurement of Technical Efficiency and Its Determinants in Wheat Production : The Case of Smallholder

Farmers in Wogidi District, South Wollo Zone Ethiopia. Journal of Food Science and Quality Management 18:12.

Dicko MH, Gruppen H, Traore AS, Voragen AGJ, Van Berkel WJH (2006). Sorghum grain as human food in Africa: relevance of starch content and amylase activities. African Journal of Biotechnology 5(5):384-395.

Dube L, Musara JP, Bediako J, Munyati VT (2014). Marketing Proclivity, Constraints and Opportunities in the Sorghum Based Stockfeed Value Chain in Lusitu, Zambia. Journal of Agriculture, Economics and Rural Development 2(3):110-122

FAO, IFAD, WFP (2014). Strengthening the enabling environment for food security and nutrition, Rome, FAO, 2014.

FAOSTAT (Food and Agriculture Organization of the United Nations statistics) (2018). Food and Agriculture Organization of the United Nations, Roma, Italy.

Fischer HW, Reddy LN, Rao MS (2016). Can more drought resistant crops promote more climate secure agriculture ? Prospects and challenges of millet cultivation in Ananthapur, Andhra Pradesh. World Development Perspectives 2:5-10.

Ghimire RG, Wen-chi HU, Hrestha RBS (2015). Factors Affecting Adoption of Improved Rice Varieties among Rural Farm Households in Central Nepal. Rice Science 22(1):35-43.

GoK (Government of Kenya) (2009). Kenya National Bureau of Statistics (KNBS). Statistical Abstract, 2009, Nairobi.

Govender L, Pillay K, Siwela M, Modi A, Mabhaudhi T (2017). Food and Nutrition Insecurity in Selected Rural Communities of KwaZulu-Natal, South Africa - Linking Human Nutrition and Agriculture. International Journal of Environmental Research and Public Health 14(1):1-21.

Grovermann C, Umesh KB, Qui S, Kumar BG, Sakamma S, Moakes S (2018). The Economic Reality of Underutilised Crops for Climate Resilience, Food Security and Nutrition: Assessing Finger Millet Productivity in India. Agriculture 8(9):131.

Hadebe ST, Modi AT, Mabhaudhi T (2017). Drought Tolerance and Water Use of Cereal Crops : A Focus on Sorghum as a Food Security Crop in Sub-Saharan Africa. Journal of Agro Crop Science 203:177-191.

Hassan AT (2015). Economics of Sorghum Production under Traditional Farming System in Nyala Governate of South Darfur State. Sudan. ARPN Journal of Science and Technology 5(2):74-79.

Hiron M, Rubene D, Mweresa CK, Ajamma YU, Owino EA, Low M (2014). Crop damage by granivorous birds despite protection efforts by human bird scarers in a sorghum field in western Kenya. Ostrich 85(2):153-159.

Hutcheson GD, Sofroniou N (1999). The multivariate social scientist: Introductory statistics using generalized linear models. Sage.

Karanja D, Kamau C, Muthoni L, Kaguma A, Kariuki CW, Kavoi J, Kega V (2009). Improving food security and income of farmers in ASALs through commercialization of gadam sorghum project. Unpublished. KARI-Kaumani, Machakos, Kenya.

Katundu BMA, Mhina ML, Arbogast G, Kumburu NP (2014). Socio-Economic Factors Limiting Smallholder Groundnut Production in Tabora Region.

Kilambaya D, Witwer M (2013). Analysis of incentives and disincentives for sorghum in Kenya. Technical notes series, MAFAP, FAO, Rome. http://www.fao.org/fileadmin/templates/mafap/documents/technical_notes/KENYA/KENYA_Technical_Note_SORGHUM_EN_Feb2013.pdf

Kloss M, Petrick M (2018). The productivity of family and hired labour in EU arable farming (No. 174). Discussion Paper, Leibniz Institute of Agricultural Development in Transition Economies.

Mabhaudhi T, Chimoyo PGV, Modi AT (2017). Status of Underutilised Crops in South Africa : Opportunities for Developing Research Capacity. Sustainability 9(9):1569.

Mabhaudhi T, O'Reilly P, Walker S, Mwale S (2016). Opportunities for underutilised crops in Southern Africa's post-2015 development agenda. Sustainability 8(4):1-16.

Maddala GS, Lahiri K (1992). Introduction to econometrics (Vol. 2). New York: Macmillan.

Masese MA, Gor C, Mukhebi A, Abuya J (2018). Analysis of agricultural extension service agents information. African Journal of Business and Industry 1(4):212–224.

Massawe FJ, Mayes S, Cheng A, Chai HH, Cleasby P, Symonds R, Azam-Ali SN (2015). The Potential for Underutilised Crops to Improve Food Security in the Face of Climate Change. Procedia Environmental Sciences 29:140-141.

Mavhura E, Manatsa D, Mushore T (2015). Adaptation to drought in arid and semi-arid environments: Case of the Zambezi Valley, Zimbabwe. Journal of Disaster Risk Studies 7(1):1-7.

Mitaru NB, Mgonja AM, Rwomushana I, Opio F (2012). Integrated sorghum and millet sector for improved livelihoods in ECA. Proceedings of the ECARSAM Stakeholders' Conference, 20th -22nd November, 2006, Dar-es-Salaam, Tanzania.

Mmbando FE, Baiyegunhi LJS (2017). Socio-economic and Institutional Factors Influencing Adoption of Improved Maize Varieties in Hai District, Tanzania. Journal of Human Ecology 53(1):49-56.

MoALF (Ministry of Agriculture, Livestock and Fisheries) (2016). Climate Risk Profile Siaya. Kenya County Climatic Risk Profile Series: The Kenya Ministry of Agriculture, Livestock and Fisheries, Nairobi, Kenya.

Mugera AW, Langemeier MR, Ojede A (2016). Contributions of Productivity and Relative Price Changes to Farm-level Profitability Change. American Journal of Agricultural Economics 98(4):1210-1229.

Mukarumbwa P, Mushunje A (2010). Potential of sorghum and finger millet to enhance household food security in zimbabwe' s semi-arid regions: A Review. Contributed Paper Presented at the Joint 3rd African Association of Agricultural Economists (AAAE) and 48th Agricultural Economists Association of South Africa (AEASA) Conference, Cape Town, South Africa, September 19-23.

Mundia CW, Secchi S, Akamani K, Wang G (2019). A Regional Comparison of Factors Affecting Global Sorghum Production: The Case of North America, Asia and Africa's Sahel. https://doi.org/10.3390/su11072135

Mutisya DL, Karanja DR, Kisilu RK (2016). Economic advantage of sorghum harvest at soft dough grain stage to prevent bird damage. Cogent Food and Agriculture 2(1):1259141.

Muui CW, Muasya RM, Kirubi DT (2013). Baseline Survey on factors affecting sorghum production and use in Eastern kenya. African Journal of Food, Agriculture, Nutrition and Development 13(1):7339-7357.

Mwadalu R, Mwangi M (2013). The potential role of sorghum in enhancing food security in semi-arid eastern Kenya: A review. Journal of Applied Biosciences, pp. 5786-5799.

Naik A, Teggelli RG, Ahamed Z, Patil DH (2016). Yield gap analysis of sorghum through front line demonstrations in Kalaburagi region of northern Karnataka. Research in Environment and Life Sciences 9(5):597-598.

Naim AM, Elobeid HA, Ahmed AE (2017). Factors affecting technical efficiency of sorghum production in New Halfa Agricultural production corporation, Sudan. Academic Journal of Life Sciences 3:1-5.

Obasi PC, Ukewuihe IS (2013). Factors Affecting Agricultural Productivity among Arable Crop Farmers in Imo State, Nigeria. American Journal of Experimental Agriculture 3(2):443-454.

Obiero EO (2013). Social economic factors affecting farm yield in Siaya District, Siaya County, Kenya. (Master Thesis) University of Nairobi, Kenya.

Ogeto RM, Cheruiyot E, Mshenga P, Onyari CN (2013). Sorghum production for food security : A socio- economic analysis of sorghum production in Nakuru County, Kenya. African Journal of Agricultural Research 8(47):6055-6067.

Okuyama Y, Maruyam A, Takagaki M, Kikuchi M (2017). Technical efficiency and production potential of selected cereal crops in Senegal. Journal of Agriculture and Rural Development in the Tropics and Subtropics 118(2):187-197.

Ombuki C (2018). Factors Influencing Maize Production in Rural Kenya : Case of Kisii County. International Journal of Arts and Commerce, 7(6):47–56.

Orr A, Mwema C, Mulinge W (2013). The value chain for sorghum beer in Kenya. CRISAT, Nairobi, KARI, Nairobi, Kenya. I. Series Paper P. 16.

Orr A, Mwema C, Gierend A, Nedumaran S (2016). Sorghum and Millets in Eastern and Southern Africa: Facts, Trends and outlook.

Pingali P (2015). Agricultural policy and nutrition outcomes - getting beyond the preoccupation with staple grains. Journal of Food Science 7:583-591.

Pradhan P, Fischer G, Velthuizen HV, Reusser DE (2015). Closing Yield Gaps : How Sustainable Can We Be ? pp. 1-18. https://doi.org/10.1371/journal.pone.0129487
Ravallion M, Chen S, Sangraula P (2007). New evidence on the urbanization of global poverty: World Bank Policy Research Working Paper 4199 (Vol. 33).
Recha CW (2018). Local and regional variations in conditions for agriculture and food security in Kenya. Njoro, Kenya.
Rezig M, Sahli A, Ben JF, Harbaoui Y (2010). Adopting intercropping system for potatoes as practice on drought mitigation under Tunisian conditions. Agronomico Mediterraneo de Zaragoza: Zaragoza, Spain 334(95):329-334.
Rutherford A (2001). Introducing ANOVA and ANCOVA: a GLM approach. Sage.
Sebastian K (2009). Agro-ecological zones of Africa. International Food Policy Research Institute: Washington, DC, USA. https://doi.org/10.7910/DVN/HJYYTI
Suvedi M, Ghimire R, Kaplowitz M (2017). Farmers' participation in extension programs and technology adoption in rural Nepal : a logistic regression analysis. Journal of Agricultural Education and Extension 23(4):351-371.
Teferi ET (2013). Adoption of improved sorghum varieties and Farmer's varietal trait preference, North Wolo Zone, Ethiopia.
Urassa JK (2017). Factors influencing maize crop production at household levels: A case of Rukwa Region in the southern highlands of Tanzania. African Journal of Agricultural Research 10(10):1097-1106.
Zalkuwi JW, Dia YZ, Dia RZ (2015). Analysis of Economic Efficiency of Maize Production in Ganye local Government Area, Adamawa state, Nigeria 2(7):1-9.

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Gender roles and constraints in the aquaculture value chain in Western Kenya

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Aquaculture plays a critical role in food and nutrition security, economic empowerment and creation of employment opportunities for millions of people. However, the benefits from aquaculture are not evenly distributed between men and women due to gender-based constraints which limit maximum returns. The present study investigated gender roles and constraints in the aquaculture value chain in Western Kenya. A household survey was conducted among 384 randomly selected farmers using structured questionnaires in three counties in Western Kenya. Results of the study reveal gender participation at different nodes of the value chain with women representation being low (32%) compared to men (68%). Gender based constraints affecting participation and benefits include access to productive resources and start-up capital and discriminatory gender norms which limit women participation and financial returns. Therefore, abolishing these constraints is imperative in increasing production for development and social wellbeing of not only women but the entire household, community and the nation at large.

Key words: Aquaculture value chain, gender roles, constraints, social norms, poverty, wellbeing.

INTRODUCTION

The State of World Fisheries and Aquaculture (FAO, 2018) draws special attention to the critical importance of fisheries and aquaculture for food and nutrition security, as well as employment for millions of people, many of whom struggle to maintain reasonable livelihoods. As the fastest-growing food-producing sector in the world, global aquaculture production grew at an average annual rate of 6.6% since 1995 (FAO, 2017). This positive shift is expected to continue; to meet the food and nutrition security, employment, and provide economic empowerment to the ever-growing population (FAO, 2018). For the past decade, Kenya's policy and legal

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frameworks have recognized aquaculture as one of the flagship projects to spur inclusive economic development. Demand for fish is increasing rapidly, driven by population and income growth, increased awareness of health benefits of fish consumption and changes in lifestyle and consumer preferences (Githukia et al., 2014; Obiero et al., 2019). However, fish supply lags, owing to declining natural fish stocks which necessitated increased fish imports in Kenya, especially Nile tilapia. Aquaculture is the most suitable alternative to capture fisheries to produce fish given the changes in climate, while also delivering co-benefits for environmental sustainability, nutrition and livelihoods (Munguti et al., 2017). Therefore, sustainable intensification of aquaculture will serve to fill the ever-widening fish demand-supply gap (Munguti et al., 2017).

However, the benefits from aquaculture are not evenly distributed between men and women due to differences in endowments and constraints associated with access to factors of production (Harrison, 1995; Ndanga et al., 2013; Kruijssen et al., 2018). The aquaculture sector is often considered a male domain because of the high levels of investment and the adoption of new technology associated with its development (Kumar et al., 2018). While fisheries and aquaculture industry empower women and contribute to gender equity; however, their role has largely been unrecognized (HLPE, 2014). Women occupy a central place in the aquaculture sector by virtue of them being at home most of the time, which unfortunately makes fish farming to be assumed as an extension of domestic duties, and therefore unrecognized and unrewarded (Ndanga et al., 2013). It is widely acknowledged that women are engaged in aquaculture in myriad ways, contributing significantly to the overall well-being of households; but the women themselves often get very little benefits in return due to deep-rooted gender disparities in social, cultural and economic spheres (Medard et al., 2001; Harrison et al., 2016).

Besides, women face stiffer constraints compared to men when it comes to access to factors of production which are often owned by men as head of the household and therefore the sole decision-makers (KMAP, 2016; Rutaisire et al., 2010). This limitation further constrains women's ability to access credit facilities since these assets form collateral. As a result, women's contribution is not commensurate to the benefits they enjoy from fish farming. Persistent differences and disparities between men and women can result in overburdening women with too much work but fewer benefits from the same with negative implications for the family and the whole society (Williams, 2000). This is especially true when women are constrained by inequality and discrimination (van Eerdewijk et al., 2017). Moreover, uneven access to factors of production and unequal distribution of benefits between genders means that aquaculture development does not benefit the whole community as expected (Ndanga et al., 2013). Considering the roles played by women, it is impossible to imagine the aquaculture sector without their incorporation (FAO, 2012).

Weeratunge et al. (2012) and FAO (2013) emphasized the importance of eliminating hurdles limiting women's control over access to assets and gender norms to attain gender equality. Additionally, Schumacher (2014) and Kruijssen et al. (2018) proposed a gender perspective in value chain analysis to mitigate the gender differences in aquaculture and increase production and returns. This entails an assessment of roles performed by men and women, and how they relate with each other; which dictates the possibilities of counteracting constraints and reaping maximum benefits from the venture. This suggestion is envisaged to encourage women's participation in the aquaculture value chain since they complement men for improved productivity and income, as well as promote gender equity (HLPE, 2014). Consequently, addressing inequalities in gender by exposing women to equal access to resources and opportunities like men increases farm production and raises agricultural output which is beneficial to the entire family (Gallant, 2019; FAO, 2011; Weeratunge-Starkloff and Pant, 2011). Since gender equality is enshrined in the Sustainable Development Goal (SDG 5) of the United Nations as a crucial target to be met by 2030 (FAO, 2011), its achievement in agriculture is indispensable (Me-Nsope and Larkins, 2015).

The present study aims to investigate gender roles and constraints in the aquaculture value chain in Western Kenya, a region which registered highest aquaculture production in Kenya. Paradoxically, the region also recorded the highest rates of poverty and malnutrition (Kundu et al., 2016; Ndanga et al., 2013; Obwanga et al., 2018). The study supports the Sustainable Development Goal 2 on zero hunger and Goal 5 on gender equality. In this paper, gender refers to the social-cultural differences among men and women which determine the roles, relationships, and responsibilities, decision making power and access to resources and control over benefits (World Bank, 2012). While applying gender analysis, the study seeks to understand the dynamics and impacts of changes on both men and women and not women's issues alone (Harrison, 1995; Lwenya et al., 2006). It is widely acknowledged that incorporating a gender lens to value chain analysis is indispensable in understanding of men's and women's roles, responsibilities and constraints and increases productivity, economic benefits and quality of livelihoods (Schumacher, 2014; Me-Nsope and Larkins, 2015).

CONCEPTUAL FRAMEWORK

To acquire adequate knowledge on gendered dimensions, this study relied on two analytical approaches that is the rapid assessment approach and the Integrating Gender into Agricultural Value Chains

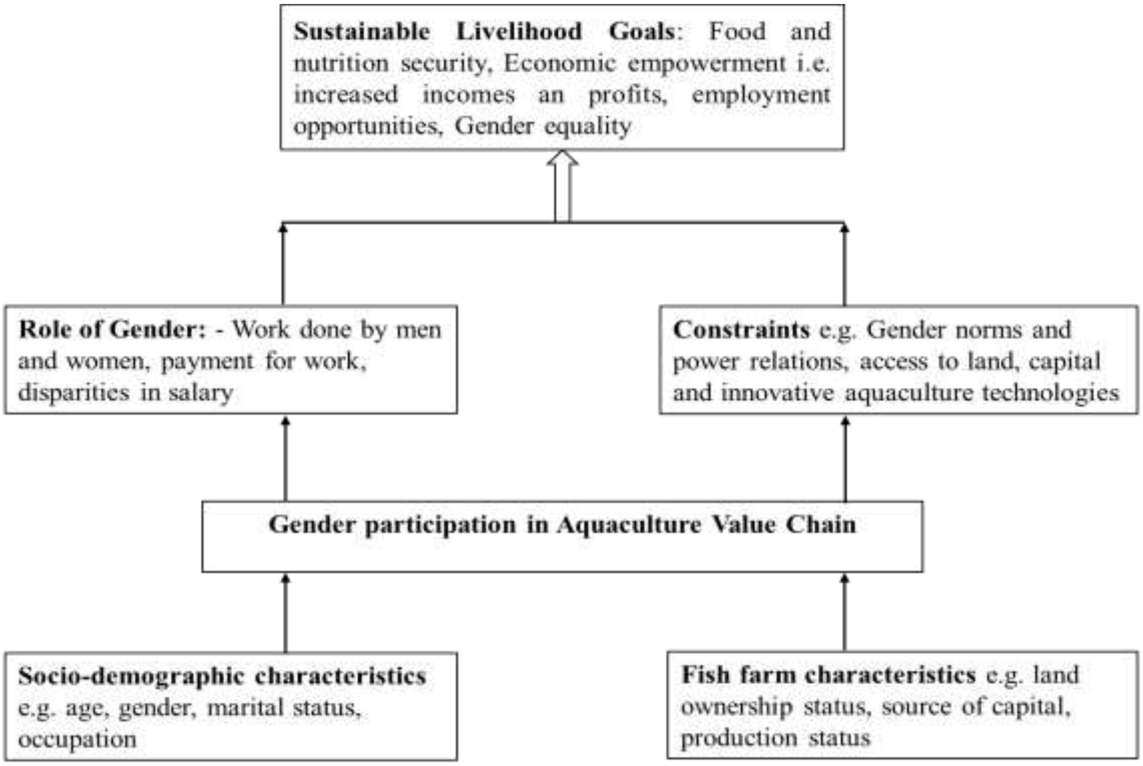

Figure 1. Conceptual framework for gender analysis in the aquaculture value chain.
Source: Modified from Kruijssen et al. (2018).

(INGIA-VC) developed by Rubin et al. (2009). These analytical tools focus on gender roles in agricultural value chains as well as their probable constraints and opportunities. For example, sociodemographic characteristics of farmers influence their roles, constraints and opportunities. Women may face stiffer constraints concerning economic factors of production since most of these factors are controlled by men in their capacity as head of the households. This challenge is also pegged on socio-cultural norms and beliefs which reduces economic options for participation and returns in the aquaculture value chain. Such gender-based differences increase inequality within households, among rural and urban inhabitants and even within countries. Farnworth et al. (2015) noted that women's contribution is rarely considered as primary factors of analysis in the aquaculture sector. Additionally, Ndanga et al. (2013) highlighted that though women participate in aquaculture; their work is mainly considered a traditional responsibility and not accorded any economic value. Coupled with low decision-making power and constraints in power relations within the households, the contribution of women is often unrecognized and unrewarded, which further constrains their productivity. This study, therefore, focuses on integrating gender participation to deliver productivity for both men and women within households.

The variables under consideration are divided into three categories, which are independent variables, intervening variables and dependent variables (Figure 1). Independent variables include the sociodemographic characteristics of fish farmers such as gender, household head and size, education level and occupation. The intervening variable is gender participation in aquaculture which affects both the independent and dependent variable. The dependent variable is gender roles which entails what men and women do in the value chain, gender constraints such as social norms which affect decision making power and benefits. The expected outcome of gender analysis is an understanding of its contribution to increased food and nutrition security among households and economic empowerment.

MATERIALS AND METHODS

This study was conducted in three counties in the Western Kenya region including Kakamega, Kisii and Homa Bay Counties. Survey design and purposive sampling method were employed to select sub-counties in each of the three counties based on fish production statistics. Respondents within each sub-county were randomly selected and a total of 384 farmers were interviewed using structured questionnaires. A reconnaissance survey was undertaken before the actual data collection to assess the type of responses expected from the field. Data were collected at the household level by trained enumerators using a digitized questionnaire in Open Data Kit (ODK), an open-source application installed on Android mobile phones. The questionnaire solicited information on socio-demographic characteristics of fish farmers, fish farm characteristics, gender roles, responsibilities, relationship, opportunities and constraints in the aquaculture value chain. The data were analysed using Stata version 13 (Stata Corp, College Station, Texas, USA) and statistical significance was considered when α = 0.05. Descriptive analyses were done by use of counts, means, median, percentages, standard deviation, and ranges to provide a better understanding of the collected data (Hejase and Hejase, 2013). This study followed the ethical considerations in research surveys involving human subjects (Alcser et al., 2011; Yin, 2009). Prior permission and consent were sought before interviews and the research team protected the rights of free will, privacy and confidentiality of respondents during the interviews and data processing.

RESULTS

Socio-demographic characteristics of the farmers

The mean age of the farmers was 49.3 years with female farmers (44.5 years) being significantly ($p < 0.001$) younger than male farmers (51.5 years) as shown in Table 1. Out of the 384 farmers interviewed, about a third (32%, n=124) were women while men formed two thirds (68%, n=260) of the sampled population. Kakamega County had the highest number of farmers (n=164, 42.7%) followed by Kisii (n=127, 33.1%) and Homa Bay (n=93, 24.2%). There was a significant difference ($p < 0.001$) in the marital status of the farmers with 82.3% being married, 7.8% single, 7.0% widowed and 2.9% separated/divorced. About 32.3% of the farmers had attained primary level of education, 36.7% had attained secondary education and 22.4% were holders of certificate and diploma certificates. A small proportion (6.7%) had an undergraduate degree and only 1% had a postgraduate degree. Forty four percent of the farmers had a household size of 4–6 members, 23.7% had 1–3 members, 27.3% had 7–10 and only 4.9% had more than 10 members. There was a highly significant difference ($p < 0.001$) in the head of the households with more male-headed households (87%) compared to female-headed households (13%).

Fish farm characteristics

The main reasons cited by farmers for venturing into fish farming included: a source of income, food for household consumption, benefit from government, and to create employment opportunities (Table 2). For female-headed households, benefit from government support was highly significant ($p = 0.001$), denoting this as a major reason for entering into fish farming while for male-headed households “creation of employment” was significant ($p = 0.02$). The main source of initial capital for fish farming was personal savings and government support with female-headed households (56%) significantly ($p = 0.011$) depending more on government support as compared to male-headed households (37.1%). More than half of the farmers had inherited land for fish farming (male, 71.2%; female, 60.5%) while the rest had purchased, leased/rented or were squatters. The mean land size utilized for fish farming was 0.21 acres (Standard Deviation, SD=0.22) for female-headed households and 0.28 acres (SD=0.8) for male-headed households but the différences were not significant. The main farmed species were Nile tilapia (female farmers, 95.2%; male farmers, 96.2%) and African catfish (37.9%, 38.9%) respectively, which were not significantly different between the gender. Most of the farmers had earthen ponds, but some owned lined and concrete ponds/tanks.

Gender roles in fish farming

Regarding occupation, 63% of the respondents were farmers, 16.7% were civil servants, 10.9% were traders, 5.2% were artisans, and 4.2% worked in diverse activities. 91.4% of the fish farmers practised fish farming on a part-time basis with 8.6% engaging in fish farming on a full-time basis (Table 3). The main activities performed alongside fish farming was agriculture (67.2%), domestic duties (40.9%), communal duties (37.2%), trading (25.3%), formal jobs (20.1%) and other income-generating activities (3.1%). There was a highly significant difference ($p < 0.001$) in gender performance of domestic duties with more female farmers (63.7%) engaged in domestic duties than male farmers (30%). 93% of farmers were engaged in fish production, 13% in feed production and 7.8% in seed production. There was a significant difference in farmer’s engagement in fish transport and processing ($p = 0.036$), trader and marketing ($p=0.044$) with more female farmers engaged in these activities compared to male farmers. The average annual income from fish farming for female farmers was KES 114,731.40 (SD = KES 102,959.10); while that of male farmers was KES 121,773.40 (SD = KES 159,052.80) but there was no significant difference between male and female farmers’ incomes. This represented 29.9% of the annual fish farm income generated by female farmers compared to 21.1% generated by male farmers which was significant ($p < 0.001$). On the other hand, the average non-fish annual income generated by female farmers was KES 219,164.50 (SD = KES 152,615.50) while that of male farmers was KES 248,607.30 (SD = KES 262,382.70) with no significant difference. Interestingly, female farmers generated 29.2% of non-farm income, while male farmers generated 20.5%; which was significantly different ($p < 0.001$). The benefits accrued from fish farming included fish for family consumption (65%), share of profits (53%), money as a gift (34%) and salary (22%). There were more women (83%) involved in unpaid work than men (17%) with 28% of farmers reporting a disparity in the amount of salary earned for work for both men and women.

Table 1. Sociodemographic characteristics of respondents by gender.

Characteristics	All n (%) n=384	Female n (%) n=124	Male n (%) n=260	p-value
Age				
Mean (SD) age	49.27 (14.09)	44.51 (13.55)	51.54 (13.80)	**<0.001**
County				
Homabay	93 (24.22)	32 (25.81)	61 (23.46)	0.263
Kakamega	164 (42.71)	58 (46.77)	106 (40.77)	
Kisii	127 (33.07	34 (27.42)	93 (35.77)	
Marital status				
Single	30 (7.81)	17 (13.71)	13 (5.00)	**<0.001**
Married	316 (82.29)	82 (66.13)	234 (90.00)	
Separated/ divorced	11 (2.86)	5 (4.03)	6 (2.31)	
Widowed	27 (7.03)	20 (16.13)	7 (2.69)	
Educational level				
Primary	124 (32.29)	42 (33.87)	82 (31.54)	0.729
Secondary	141 (36.72)	48 (38.71)	93 (35.77)	
Certificate/Diploma	86 (22.40)	27 (21.77)	59 (22.69)	
Undergraduate	26 (6.77)	6 (4.84)	20 (7.69)	
Postgraduate	4 (1.04)	0 (0)	4 (1.54)	
Others	3 (0.78)	1 (0.81)	2 (0.77)	
Household size				
1 to 3	91 (23.70)	38 (30.65)	53 (20.38)	**0.042**
4 to 6	169 (44.01)	56 (45.16)	113 (43.46)	
7 to 10	105 (27.34)	27 (21.77)	78 (30.00)	
10 and above	19 (4.95)	3 (2.42)	16 (6.15)	
Head of household				
Mother	50 (13.02)	44 (35.48)	6 (2.31)	**<0.001**
Father	334 (86.98)	80 (64.52)	254 (97.69)	

Gender constraints in fish farming

Norms and perceptions

In terms of social norms, 13.5% of respondents reported the existence of social norms in fish farming concerning what women and men are allowed to do that is in form of labor, who they can interact with and their mobility (Table 4). Almost equal number of female farmers (60.5%) and male farmers (60.8%) reported women to be experiencing mobility challenges due to their domestic engagements. However, there was a highly significant difference ($p < 0.001$) in female (82.9%) compared to male (66%) farmers who reported women to be responsible for food security. In addition, there was a significant difference ($p = 0.001$) in female (37.1%) and male (55.4%) farmers who reported men to be the main income earners. Compared to women, men significantly ($p < 0.001$) made decisions in the management of fish farm enterprises. Among the reasons that hinder women from accessing and benefiting from the aquaculture sector included unbalanced gender norms, power relations, capital, education, and confidence levels. Gender norms ($p=0.020$) and power relations ($p=0.003$) were significant between gender. More male farmers had significantly ($p=0.009$) more control over incomes and profits earned from fish farming as compared to female farmers. Also, male farmers had a highly significant ($p < 0.001$) returns from fish farming as compared to female farmers.

Table 2. The distribution of fish farm characteristics by gender.

Fish farm characteristics	Gender of the respondent			Gender of the household head		
	Female	Male	p-value	Female	Male	p-value
	n (%)	n (%)		n (%)	n (%)	
	n=124	n=260		n=50	n=334	
Reasons for venturing into fish farming						
Source of income	111 (89.52)	241 (92.69)	0.292	45 (90.00)	307 (91.92)	0.648
Food for household consumption	67 (54.03)	139 (53.46)	0.916	27 (54.00)	179 (53.59)	0.957
Government support programmes	46 (37.10)	73(28.08)	**0.020**	18 (36.00)	101 (30.24)	**0.001**
Create employment	50 (40.32)	134 (51.54)	**0.048**	20 (40.00)	164 (49.10)	**0.020**
Others	1 (0.81)	12 (4.62)	0.069	0 (0)	13 (3.89)	0.390
Sthece of initial capital						
Government support	59 (47.58)	93 (35.77)	**0.027**	28 (56.00)	124 (37.13)	**0.011**
Bank loan or microfinance	9 (7.26)	17 (6.54)	0.793	5 (10.00)	21 (6.29)	0.330
Grants from NGO/CBO	23 (18.55)	40 (15.38)	0.434	6 (12.00)	57 (17.07)	0.367
Personal savings	89 (71.77)	192 (73.85)	0.668	33 (66.00)	248 (74.25)	0.219
Friends/relatives	13 (10.48)	27 (10.38)	0.976	4 (8.00)	36 (10.78)	0.549
Others	0 (0)	3 (1.15)	0.554	0 (0)	3 (0.90)	1.000
Land ownership status						
Purchased	36 (29.03)	56 (21.54)	0.114	11 (22.00)	81 (24.25)	0.576
Inheritance	75 (60.48)	185 (71.15)		33 (66.00)	227 (67.96)	
Lease/rent	9 (7.26)	16 (6.15)		5 (10.00)	20 (5.99)	
Squatter	3 (2.42)	1 (0.38)		1 (2.00)	3 (0.90)	
Others (specify)	1 (0.81)	2 (0.77)		0 (0)	3 (0.90)	
Total land area for fish farming, m^2						
Mean (SD)	1315.67 (4937.19)	1001.49 (1420.90)	0.344	867.08 (893.35)	1138.25 (3236.02)	0.557
Fish species produced						
Tilapia	118 (95.16)	250 (96.15)	0.649	50 (100)	318 (95.21)	0.243
Catfish	47 (37.90)	101 (38.85)	0.859	16 (32.00)	132 (39.52)	0.308
Production facilities						
Earthen ponds	102 (82.26)	225 (86.54)	0.270	40 (80.00)	287 (85.93)	0.272
Liner ponds	37 (29.84)	63 (24.23)	0.242	18 (36.00)	82 (24.55)	0.085
Concrete ponds/tanks	9 (7.26)	17 (6.54)	0.793	4 (8.00)	22 (6.59)	0.761

Constraints by access to and control over assets and resources

On the other hand, 52.3% of the male farmers owned the main factors of production for example farm equipment, production facilities and land compared to 36.3% of the female farmers which was significantly different ($p = 0.001$) as presented in Table 5. In addition, there was a highly significant difference ($p < 0.001$) in land ownership, with male farmers (56.9%) owning more land than female farmers (31.5%). More than a quarter of fish farmers sourced capital from friends/relatives (females 29%, males 37%) while the rest accessed capital from microfinance, banks or borrowed from their spouses with more females (16%) depending on their spouses than males (7%). Farmers faced challenges in accessing loan

Table 3. Understanding the distribution of labthe by gender in aquaculture: Roles and perceptions.

Labour division	Overall n (%)	Female n (%)	Male n (%)	p-value
Main occupation:				
Civil servant	64 (16.67)	23 (18.55)	41 (15.77)	0.118
Farmer	242 (63.02)	75 (60.48)	167 (64.23)	
Trader	42 (10.94)	17 (13.71)	25 (9.62)	
Artisan	20 (5.21)	2 (1.61)	18 (6.92)	
Other	16 (4.17)	7 (5.65)	9 (3.46)	
Do you work in the fish farm on a full-time or part-time basis?				
Full-time	33 (8.59)	9 (7.26)	24 (9.23)	0.519
Part-time	351 (91.41)	115 (92.74)	236 (90.77)	
What other activities are performed alongside fish farming?				
Domestic duties	157 (40.89)	79 (63.71)	78 (30.00)	**<0.001**
Community duties	143 (37.24)	51 (41.13)	92 (35.38)	0.276
Formal jobs	77 (20.05)	31 (25.00)	46 (17.69)	0.094
Other farm activities	258 (67.19)	78 (62.90)	180 (69.23)	0.217
Trading	97 (25.26)	32 (25.81)	65 (25.00)	0.865
Others	12 (3.13)	2 (1.61)	10 (3.85)	0.351
In which aquaculture sector do you work?				
Feed production	50 (13.02)	18 (14.52)	32 (12.31)	0.548
Seed production	30 (7.81)	10 (8.06)	20 (7.69)	0.899
Fish production	357 (92.97)	117 (94.35)	240 (92.31)	0.463
Transport and Processing	111 (31.09)	45 (38.46)	66 (27.50)	**0.036**
Trader/Marketing	165 (46.22)	63 (53.85)	102 (42.50)	**0.044**
Mean (SD) annual fish farm income	119499.8 (143257.1)	114731.4 (102959.1)	121773.9 (159052.8)	0.653
Percentage generated by women	43.13 (30.34)	69.40 (29.94)	30.60 (21.16)	**<0.001**
Mean (SD) non-farm annual income	239099.7 (232863.8)	219164.5 (152615)	248607.3 (262382.7)	0.247
Percentage generated by women	45.87 (26.51)	63.27 (29.18)	37.58 (20.52)	<0.001
What are women paid for working in the fish farm?				
They get a salary	83 (21.61)	35 (28.23)	48 (18.46)	0.030
They get a share of profits	205 (53.39)	70 (56.45)	135 (51.92)	0.406
They get money as a gift (not regularly)	132 (34.38)	48 (38.71)	84 (32.31)	0.217
They get fish to eat	251 (65.36)	76 (61.29)	175 (67.31)	0.247
Others	3 (0.78)	1 (0.81)	2 (0.77)	1.000
Are more women or more men involved in unpaid work?				
Women	320 (83.33)	111 (89.52)	209 (80.38)	0.025
Men	64 (16.67)	13 (10.48)	51 (19.62)	
Is there a disparity in the amount of salary earned for work by men and women?				
Yes	106 (27.68)	32 (25.81)	74 (28.57)	0.571
No	277 (72.)	92 (74.19)	185 (71.43)	

Table 4. Distribution of perceived gender norms and distribution of benefits by gender.

Characteristics	Overall n (%) n=384	Female n (%) n=124	Male n (%) n=260	p-value
Is there existence of gender norms (in terms of Labour, interaction, and mobility)?				
Yes	52 (13.54)	22 (17.74)	30 (11.54)	0.097
Women's mobility limited compared to men's				
Yes	233 (60.68)	75 (60.48)	158 (60.77)	0.957
Women mainly responsible for food security?				
Yes	252 (71.59)	97 (82.91)	155 (65.96)	<0.001
Are men the main earners				
Yes	190 (49.48)	46 (37.10)	144 (55.38)	0.001
Between men and women, who make more decisions in the fish farm?				
Women	85 (22.14)	63 (50.81)	22 (8.46)	<0.001
Men	299 (77.86)	61 (49.19)	238 (91.54)	
Reason that hamper women from accessing and benefitting from the aquaculture sector?				
Gender norms	124 (32.29)	50 (49.32)	74 (28.46)	0.020
Power relations	200 (52.08)	78 (62.90)	122 (46.92)	0.003
Capital	246 (64.06)	88 (70.97)	158 (60.77)	0.051
Education	146 (38.02)	52 (41.94)	94 (36.15)	0.275
Confidence	168 (43.75)	50 (40.32)	118 (45.38)	0.350
Others	12 (3.13)	4 (3.23)	8 (3.08)	1.000
Do you have control over the income and profits earned from fish farming?				
Yes	337 (87.76)	101 (81.45)	236 (90.77)	0.009
Between men and women who gets more returns from fish farming?				
Women	103 (26.82)	65 (52.42)	38 (14.62)	<0.001
Men	281 (73.18)	59 (47.58)	222 (85.38)	

facilities for fish farming because of repayment challenges and lack of collateral. However, these reasons were not significant between genders. Respondents proposed several mechanisms to promote gender participation in fish farming which included access to aquaculture technologies, best management practices and focus on dissemination of entrepreneurial and technical skills through extension and advisory services.

DISCUSSION

Even though women comprised a third of the fish farmers and were relatively younger than their male counterparts, the majority of the farmers' households were male-headed. The findings corroborate with studies by Obiero et al. (2019), Ole-Moiyoi (2017) and KMAP (2016) recognizing the majority of fish farming households to be male-headed. However, women play a crucial role in the aquaculture industry, since they make a significant contribution at different nodes of the value chain (Ndanga et al., 2013; Weeratunge and Snyder, 2009). In particular, they play a crucial role in the control of production (Brugere and Williams, 2017) and food and nutrition security (Obwanga and Lewo, 2017). However, this trend works against women especially in terms of decision making, benefits sharing and power relations within the households (Kruijssen et al., 2018). Ndanga et al. (2013)

Table 5. Constraints by access to and control over assets and resources.

Characteristics	Gender of the respondent			Gender of household head		
	Female	Male	p-value	Female	Male	p-value
	n (%)	n (%)		n (%)	n (%)	
Ownership of farm equipment and production facilities?						
Yourself	45 (36.29)	136 (52.31)	**0.001**	31 (62.00)	150 (44.91)	0.017
Spouse	16 (12.90)	8 (3.08)		3 (6.00)	21 (6.29)	
Household	59 (47.58)	107 (41.15)		15 (30.00)	151 (45.21)	
Informal group	2 (1.61)	4 (1.54)		1 (2.00)	5 (1.50)	
Institution (school)	2 (1.61)	5 (1.92)		0 (0)	7 (2.10)	
Land ownership						0.174
Yourself	39 (31.45)	148 (56.92)	**<0.001**	30 (60.00)	157 (47.01)	
Spouse	32 (25.81)	7 (2.69)		6 (12.00)	33 (9.88)	
Household	49 (39.52)	95 (36.54)		12 (24.00)	132 (39.52)	
Informal group	3 (2.42)	4 (1.54)		2 (4.00)	5 (1.50)	
Institution	1 (0.81)	6 (2.31)		0 (0)	7 (2.10)	
Source of capital						0.106
Microfinance	30 (21.19)	66 (25.38)	0.034	11 (22.00)	85 (25.45)	
Banks	20 (16.13)	32 (12.31)		9 (18.00)	43 (12.87)	
Friends/Relatives	37 (29.84)	98 (37.69)		24 (48.00)	111 (33.23)	
Borrow from spouse	20 (16.13)	18 (6.92)		4 (8.00)	34 (10.18)	
Others	17 (13.71)	46 (17.69)		2 (4.00)	61 (18.26)	
How easy it is to get a loan for your fish farming business?						0.034
Extremely easy	5 (4.03)	6 (2.31)	0.769	1 (2.00)	10 (2.99)	
Easy	19 (15.32)	40 (15.38)		3 (6.00)	56 (16.77)	
Fairly easy	21 (16.94)	52 (20.00)		6 (12.00)	67 (20.06)	
Difficult	46 (37.10)	102 (39.23)		22 (44.00)	126 (37.72)	
Very difficult	33 (26.61)	60 (23.08)		18 (36.00)	75 (22.46)	
If difficult or very difficult, reasons						0.065
Lack of collateral	44 (35.48)	81 (31.15)	0.397	24 (48.00)	101 (30.24)	
Inability to service the loan	63 (50.81)	113 (43.46)	0.177	30 (60.00)	146 (43.71)	
Other (specify)	6 (4.84)	18 (6.92)	0.430	4 (8.00)	20 (5.99)	
Have access to aquaculture technologies	96 (77.42)	186 (71.54)	0.222	38 (76.00)	244 (73.05)	
Have access to best management practices	90 (72.58)	181 (69.62)	0.551	37 (74.00)	234 (70.06)	
Have access to entrepreneurial and technical skills through extension services	68 (54.84)	141 (54.23)	0.911	28 (56.00)	181 (54.19)	0.012

and Harrison (1995) also echoed the same sentiments noting that men have easier access to productive resources compared to women. Based on broader studies on women engagement in the agricultural value chain, Chete (2019) found a significant contribution of women in the value chain. Results indicate the majority of farmers had primary and secondary level of education, which concurs with Ole-Moiyoi (2017). This gives them basic knowledge on matters related to fish production and marketing. Moreover, they understand the basic information on extension service delivery to update themselves with vital knowledge on technical and entrepreneurial skills (Obiero et al., 2019). Majority of the farmers had a household size of 4-6 members which agrees with a report by KMAP (2016) reporting a similar family size in Kenya. However, this is slightly higher than the 3.9 figure reported in the just concluded census in Kenya (KPHC, 2019).

Overall, most farmers were engaged in fish farming for income generation and food for household consumption which are very important for livelihood improvement (Phillips et al., 2016; Edwards, 2000; Okechi, 2004).

These findings also agree with previous studies by Obiero et al. (2019) and Kiumbuku et al. (2013). Furthermore, results revealed that personal savings and government support were the major sources of capital by both gender with female farmers having a higher reliance on government support. Noteworthy, the dependence on government support for female-headed households might be attributed to constraints of capital sources hindering them from the entry into fish farming. The government initiative through the Economic Stimulus Program (ESP) in the year 2009 to 2012, resulted in an upward and significant growth from 4,218 metric tonnes (MT) in 2006 to a peak of 24,096 in 2014 (Munguti et al., 2017; Obiero et al., 2019). Besides, there was an increase in the value of aquaculture product from Kshs 1.041 billion in 2009 to about Kshs 4.634 billion (US$56 million) in 2012 (SDF, 2012). However, when the support ended, the sector registered a decrease in production to about 14,952 MT in 2016 (KMFRI, 2017). Therefore, the sustainability of aquaculture in the country was challenged after the government support through ESP ended as argued by Amankwah et al. (2016). In this regard, there should be mechanisms by farmers to ensure sustainability and reduce the overdependence on government support.

Furthermore, the present study findings reveal that land was mainly inherited with more male farmers having inherited land compared to their female counterparts. Land inheritance favours the male members and is an old discriminatory tradition and a constraint for women hindering gender equity and productivity. Male headed households had slightly more land than female-headed households which corroborates a study by KMAP (2016) reporting that most of the land for fish farming in Kenya was owned by men. Elsewhere, Ajani (2008) reported that patriarchal arrangements in Nigeria favours men at the expense of women by allocating them more productive land. Sexsmith and Speller (2017) and Chete (2019) posited that women possessed smaller pieces of land compared to men which were attributed to lack of statutory land rights and patriarchal land systems. This societal trend of favouring men as landowners compared to women has been perpetuated over generations in many communities and constitute bias against women. Earthern ponds were the main production units while Nile tilapia was the main cultured species, an observation earlier reported by Munguti et al. (2014), Mucai et al. (2011) and Ngugi et al. (2018).

Farming is the main economic activity of the sampled population, confirming the importance of the agricultural sector in Kenya as essential sources of economic growth, employment, poverty reduction and food security for more than 80 per cent of the Kenyan population (FAO, 2010; UNDP, 2018). Female farmers were mainly engaged in domestic duties characterized by multiple and simultaneous activities within the household which takes much of their productive time as compared to male farmers. Additionally, women were engaged in fish transport, processing and marketing as opposed to men who mainly produced fish yet most of their work was unpaid. This study corroborates findings by Ndanga et al. (2013), Olufayo (2012), Rutaisire et al. (2010) and Lwenya and Abila (2000), Abila et al. (2009) and Ikiara (1999). The significant contribution of women is important, especially when examining the effect of gender relations on aquaculture production since it is impossible to imagine the aquaculture sector without women especially on the terminal end of the value chain (Harrison, 2000).

Ndanga et al. (2013) demonstrated that though women are actively involved in different nodes of the aquaculture value chain, much of their work is unpaid and unrecognized and mostly assumed to be an extension of domestic duties. Their active participation geared towards improving food and nutrition security and economic wellbeing of households (Lewis, 1997; Edwards, 2000; Genschick et al., 2018) cannot be overemphasized. Therefore, failure to remunerate women for their productive roles is not just subordinative but diminishes productivity, compromises production and circumscribes sustainable livelihoods (Chete, 2019). To achieve economic growth and food security, women efforts have to be fully recognized and rewarded which requires concerted efforts and support at the household, community and national level. The implementation of ESP spurred aquaculture growth in Kenya (Nyandat and Owiti, 2013) presenting many opportunities under the prevalent growth strategies for women to participate in aquaculture (Ndanga et al., 2013). The results are consistent with Cohen et al. (2016) who found that women's labour demands continued to escalate as livelihood activities diversify. Additionally, de Haas (2009) noted that changes in gender roles among households are correlated with improved livelihood pursuits.

Female and male respondents agreed that there were social norms, interaction and mobility challenges which mainly affected women and were associated with domestic responsibilities. As such, women capacity to pursue a broader range of livelihood activities and to attend training was limited by physical mobility restraints. The same sentiments were echoed by Lawless et al. (2019) who noted serious gender norms restricting women from leaving the households in the Solomon Islands since their husbands could not undertake domestic duties because it was against customary expectations. Also, this was a way of challenging existing power relations and a high form of disrespect for their husbands (Boudet et al., 2013) and could increase tension in relationships. Family responsibilities reduce women availability in meetings and limit their participation as they carry out other domestic activities. Gender equity in aquaculture can be enhanced through engaging both men and women in a gender transformative approach entailing shifting inequitable norm-based constraints which hinder full participation, production and benefits.

Access to factors of production for example land was a greater constraint for women than for men and this, in turn, limited women's ability to access loan because they lacked collateral and financial leverage. This is a double tragedy and hinders women capacity to improve productivity geared towards better livelihoods and development (KIT, Agri-ProFocus, IIRR, 2012; Ndanga et al., 2013; Harrison, 1995). In many communities, land inheritance is tagged to discriminatory laws and patriarchal land systems (Sexsmith and Speller, 2017) which labour men at the expense of women. For example, women own about 10-30 % of land in Africa, about 10% in Ghana, and a meagre 5% in Mali and Kenya (Doss, 2005; Deere and Doss, 2006; Chete, 2019). In Pakistan, Ecuador and Bangladesh women-headed households control smaller portions of land compared to male-headed households (FAO, 2011). Brugere and Williams (2017) and Kiumbuku et al. (2013) postulated that constraints to factors of production which are associated with gender norms limit women engagement in aquaculture with negative implication to food and nutrition security. Evidence by Ajani (2008) highlighted that patriarchal arrangements allocate bigger and more productive land to men denying women of commensurate access which is discriminatory. Moreover, Gilbert et al. (2002) and Holden et al. (2001) posit this trend to be leading to lower yields and as a result unproductive. The major constraints experienced by farmers are common in the agricultural sector as reported by Veliu et al. (2009) and Me-Nsope and Larkins (2015).

Therefore, increasing women's access to factors of production could optimize production by 20–30 per cent and raise output in developing countries by 2.5–4 per cent and at the same time reduce the number of hungry people in the world by 12–17 per cent (Peterman et al., 2010; FAO, 2010). Consequently, enhanced gender participation in aquaculture, among other issues highlighted by (FAO, 2012) could promote equitable access to and control over resources which is salient in addressing SDG 2 and SDG 5 on reducing hunger and ensuring gender equality for holistic development. This is in line with studies by Weeratunge et al. (2010) that reported a direct link between gender equity and social and economic growth.

Women also faced a challenge in accessing entrepreneurial skills and education which was associated with challenges of mobility and gender inequality as reported by Kruijssen et al. (2018). A study by the World Bank (2012) and UNDP (2018) highlighted that such gender inequality and discrimination negatively affects production and development outcomes and social wellbeing of not only women but the entire household and community at large. There is a correlation between gender inequality and increased poverty levels, poor economic growth and social wellbeing (Weeratunge et al., 2010). Besides, Gallant (2019) reported that gender norms negatively affect the decision making of women within households, increasing the burdens and reducing benefits accrued from farming.

Male farmers had more control over incomes and profits earned from fish farming as compared to female farmers. This agrees with findings by Gallant (2019), Ndanga et al. (2013) and Kruijssen et al. (2013) who noted that the benefits from fish farming are not equally accessible or distributed to the men and women who engage and depend on it. Access to credit facilities could result in significant improvements within households thereby positively influence productivity and improving livelihoods. Several authors found a positive relationship between microcredit and women's decision-making power within households and control over assets (Amin et al., 1995; Jamal, 2008) and women empowerment (Mayoux, 2006, Malhotra et al., 2002) and power relations within the households (Mizan, 1993; Kabeer, 2001). Chete (2019) noted that gender inequality in agriculture is mostly manifested in access to and control over resources and benefits. However, this study found no discrimination between the income earned for work between men and women. This study contributes to gender awareness of roles and opportunities along the aquaculture value chain. The authors recommend similar studies to be conducted in other parts of the country.

Conclusion

Both women and men's complementary roles were evident in the aquaculture value chain and production, transport and marketing. Women contribution was significant, though their representation was low as compared to their male counterparts and most of their work was unpaid. Moreover, women faced constraints associated to access to factors of production, control over income and benefits, gender norms, and mobility. Therefore, providing women with more access to factors of production and abolishing discriminatory norms and relations limiting optimum participation and benefits will raise aquaculture output, development outcomes and social wellbeing of not only women but the entire household, community and the nation at large. This is also imperative in releasing SGD 2 on zero hunger and SGD 5 on gender equality for holistic development taking in to account the link between gender equity and social and economic growth.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Abila RO, Yongo EO, Lwenya C (2009). Socio-economic baseline survey of the fishing communities of Lake Victoria, Kenya. KMFRI Technical Report No. 1/ 2009, Kisumu, Kenya.

Ajani OIY (2008). Gender dimensions of agriculture, poverty, nutrition and food security in Nigeria. Nigeria Strategy Support Program (NSSP). Background Paper No. NSSP 005, IFPRI- Abuja.

Alcser K, Benson G, Guyer H (2011). Pilot testing the SHARE quality profile. In M. Schröder (Ed.), Retrospective data collection in the survey of health, ageing and retirement in Europe. SHARELIFE Methodology. Mannheim, Germany: MEA. (pp. 37-43). Retrieved June 14. 2016, from http://www.share-project.org/uploads/tx_sharepublications/FRBMethodology_2011_Ch.5.pdf

Amankwah A, Quagrainie KK, Preckel PV (2016). Demand for improved fish feed in the presence of a subsidy: A double hurdle application in Kenya. Agricultural Economics 47:633643.

Amin R, Hill RB, Li Y (1995). Poor women's participation in credit-based self-employment: the impact on their empowerment, fertility, contraceptive use, and fertility desire in rural Bangladesh. The Pakistan Development Review 34(2):93-119.

Boudet A, Petesch P, Turk C, Thumala A (2013). On norms and agency: Conversations about gender equality with women and men in 20 countries. In Directions in development. Washington, DC: World Bank.

Brugere C, Williams M (2017). Profile: Women in Aquaculture. https://genderaquafish.org/portfolio/women-in-aquaculture/

Chete OB1 (2019). Gender and agricultural practice in developing countries: Literature review. South Asian Jthenal of Social Studies and Economics 5(2):1-11.

Cohen PJ, Lawless S, Dyer M, Morgan M, Saeni E, Teioli H, Kantor P (2016). Understanding adaptive capacity and capacity to innovate in social-ecological systems: Applying a gender lens. Ambio 45(3):S309–S321.

Deere CD, Doss C (2006). Gender and the distribution of wealth in developing countries. UNUWIDER (World Institute for Development Economics Research of the United Nations University), Research Paper No. 2006/115, UNU-WIDER, Helsinki.

de Haas H (2009). Mobility and human development. Human Development Research Paper. Vol. 01. UNDP.

Doss C (2005). The effects of intra-household property ownership on expenditure patterns in Ghana. Jthenal of African Economies 15:149-180.

Edwards P (2000). 'Aquaculture, Poverty Impacts and Livelihoods'. Jthenal of Natural Resthece Perspectives P. 56. http://www.odi.org.uk/nrp/56.html.

FAO (2018). The State of World Fisheries and Aquaculture 2018. Meeting the sustainable development goals. Rome. Licence: CC BY-NC-SA 3.0 IGO. https://creativecommons.org/licenses/by-nc-sa/3.0/igo

FAO (2017). Ending poverty and hunger by investing in agriculture and rural areas. Penang, Malaysia. http://www.fao.org/3/9-i7556e-pdf

FAO (2013). Mainstreaming gender in fisheries and aquaculture. A stock-taking and planning exercise. Final Report, Rome, Italy. P. 55. ISBN 978-92-5-107473-2

FAO (2012). The State of World Fisheries and Aquaculture, Food and Agriculture Organization of the United Nations, Rome, Italy. P. 209. ISSN 1020-5489

FAO (2011). The state of food and agriculture: Women in agriculture: Closing the gender gap for development, Rome, Italy. http://www.fao.org/publications/sofa/2010-11/en/

FAO (2010). The State of Food Insecurity in the World. Addressing food insecurity in protracted crises. Food and Agriculture Organization of the United Nations, Rome, Italy. ISBN 978-92-5-106610-2

Farnworth CR, Sultana N, Kantor P, Choudhury A (2015). Gender integration in aquaculture research and technology adoption processes: Lessons learned in Bangladesh. Penang, Malaysia: WorldFish. Working paper: 2015-17.

Gallant M (2019). Understanding gendered preferences for Climate-Smart Agriculture adoption in Malawi. Major Research Paper submitted in partial fulfilment of the requirements of the Master of Arts in International Development and Globalization. University of Ottawa, Canada.

Genschick S, Marinda P, Tembo G, Kaminski AM, Thilsted SH (2018). Fish consumption in urban Lusaka: The need for aquaculture to improve targeting of the poor. Aquaculture 492:280-289. https://www.researchgate.net/publication/324384068.

Gilbert RA, Sakala WD, Benson TD (2002). Gender analysis of a nationwide cropping system, trial survey in Malawi. African Studies Quarterly 6:1-2. Spring 2002 ISSN: 2152-2448

Githukia CM, Obiero KO, Manyala JO, Ngugi CC, Quagrainie KK (2014). Consumer perceptions and preferences of wild and farmed Nile Tilapia (*Oreochromis niloticus* L.) and African Catfish (*Clarias gariepinus* Burchell 1822) in urban centres in Kenya. International Journal of Advanced Research 2(7):694-705. http://www.jthenalijar.com

Harrison R, Leitch C, McAdam M (2016). Identity work and the development of entrepreneurial leadership: does gender matter? In: T. Nelson, K. Lewis, & C. Henry (Eds.), Global female entrepreneurship handbook. London: Routledge.

Harrison E (2000). Gender, rights and poverty issues: Lessons for the sector, Background paper for DFID/FGRP-3/ARP Workshop on practical strategies for poverty targeted research, 7-11 November, Overseas Development Institute.

Harrison E (1995). Fish and feminists. IDS Bulletin 26(3):39-47. Institute of Development Studies. 10.1111/j.1759-5436.1995.mp26003005.x

Hejase AJ, Hejase HJ (2013). Research Methods: A practical approach for business students (2nd edition). Philadelphia, PA, USA: Masadir Inc.

HLPE (2014). Food losses and waste in the context of sustainable food systems. A report by the High Level Panel of Experts on Food Security and Nutrition of the Committee on World Food Security. Rome, Italy 2014.

Holden ST, Shiferaw B, Pender J (2001). Market imperfections and profitability of land use in the Ethiopian Highlands: A comparison of selection Models with heteroskedasticity. Journal of Agricultural Economics 52(2):53-70.

Ikiara MM (1999). Sustainability, livelihoods, production and effort supply in a declining fishery: The case of Kenya's Lake Victoria Fisheries. University of Amsterdam. PhD Thesis.

Jamal H (2008). Exploring the impact of microfinance in Pakistan. Research Report No. 77. Karachi: Social Policy and Development Centre. http://www.spdc-pak.com Accessed October 28, 2019.

Kabeer N (2001). Conflicts over credit: Re-evaluating the empowerment potential of loans to women in Rural Bangladesh. World Development 29(1):63-84. https://doi.org/10.1016/S0305-750x(00)00081-4

KIT, Agri-ProFocus, IIRR (2012). Challenging chains to change: Gender equity in agricultural value chain development. KIT Publishers, Royal Tropical Institute, Amsterdam. https://www.kit.nl/gender/wp-content/uploads/publications/2008_chachacha.pdf. Accessed November 24, 2019

Kiumbuku S, Mutinda J, Bernard J (2013). Forms of gender inequalities in fish farming in Kwanza Division, Trans Nzoia County, Kenya. Research on Humanities and Social Sciences 3(15):1-9. ISSN 2222-1719.

KMAP, Kenya Market-Led Aquaculture Programme (2016). Gender impact Study Submitted by ETC East Africa to Farm Africa. P. 24.
KMFRI (2017). Kenya's Aquaculture Brief 2017: Status, Trends, Challenges and Future Outlook. Kenya Marine and Fisheries Research Institute, Mombasa, Kenya. P. 12.
KPHC (2019). Kenya Population and Housing Census. Volume I: Population by County and Sub-County, Kenya National Bureau of Statistics. November, 2019. Nairobi, Kenya.
Kruijssen F, McDougal CL, van Asseldonk IJM (2018). Gender and aquaculture value chains: A review of key issues and implications for research. Aquaculture 493:328-337.
Kruijssen F, Albert JA, Morgan M, Boso D, Siota, F, Sibiti S, Schwarz AJ (2013). Livelihoods, markets, and gender roles in Solomon Islands: Case studies from Western and Isabel Provinces. Penang, Malaysia: CGIAR Research Program on Aquatic Agricultural Systems. Project Report: AAS-2013-22. http://pubs.iclarm.net/resthece_centre/AAS-2013-22.pdf.
Kumar G, Engle C, Tucker C (2018). Factors driving aquaculture technology adoption. Journal of World Aquaculture Society 49:447-476.
Kundu R, Muchiri M, Njiru M, Nyamweya C (2016). Effect of social and economic drivers on success of small scale fish farming in Western Kenya. African Journal of Tropical Hydrobiology and Fisheries 14:29-44. https://www.researchgate.net/publication/325734727
Lawless S, Cohen P, McDougall C, Orirana G, Siota F, Doyle K (2019). Gender norms and relations: implications for agency in coastal livelihoods. Maritime Studies, https://doi.org/10.1007/s40152-019-00147-0
Lewis D (1997). Rethinking aquaculture for resthece-poor farmers: Perspectives from Bangladesh. Food Policy 22(6):533-546.
Lwenya CA, Lwenya KR, Abila R, Omwenga R (2006). Gender participation in Fisheries Management in Lake Victoria, Kenya. Kenya Marine and Fisheries Research Institute. http://oceans.org/bitstream/1834/3570/1/WLCK-266-272. Accessed January 20, 2020.
Lwenya C, Abila R (2000). The Gender role in fish processing and trading practices in Lake Victoria, Kenya. Kenya. Kenya Marine and Fisheries Research Institute. http://hdl.hanle.net/11671/1946. Accessed November 18, 2019.
Malhotra A, Schuler RS, Boender C (2002). Measuring women's empowerment as a variable in international development. Washington, DC: World Bank. https://doiI:10.1037/e597202012-004
Mayoux L (2006). Gender mainstreaming in value chain development: Experience with gender action learning system in Uganda. Enterprise Development and Microfinance Journal GALS experience Practical Action P. 15.
Medard M, Sobo F, Ngatunga T, Chirwa S (2001). Women and gender participation in the fisheries sector in Lake Victoria. 6th Asian Fisheries Forum. Kaohsiung, Taiwan. 29 November, 2001.
Me-Nsope N, Larkins M (2015). Gender analysis of the pigeon pea value chain: Case study of Malawi. Center Report Series, No. 4. Global Center for Food Systems Innovation, Michigan State University, East Lansing, Michigan, USA. ISBN: 978-0-9903005-5-7
Mizan AN (1993). Women's decision-making power in rural Bangladesh: A case study of Grameen. In: AbuWahid, (ed.) The Grameen Bank: the poverty relief in Bangladesh. Dhaka, Bangladesh: West View Press. pp. 97-126.
Mucai M, Wangila BC, Norman N (2011). Factors determining structure and development of fish farming among small scale operators in Western Kenya. In Samaki News: Aquaculture Development in Kenya towards Food Security, Poverty Alleviation and Wealth Creation 7(1):30-45.
Munguti JM, Obiero KO, Orina PS, Musa S, Mwaluma J, Mirera DO, Ochiewo J, Kairo J, Njiru JM (2017). State of Aquaculture in Kenya. Laxpress Services, Nairobi, Kenya P. 133.
Munguti JM, Ogello EO, Kim J (2014). An overview of Kenyan aquaculture: Current status, challenges and opportunities for future development. Jthenal of Fisheries and Aquatic Science 17(1):1-11.
Ndanga LZB, Quagrainie KK, Dennis JH (2013). Economically feasible options for increased women participation in Kenyan Aquaculture value chain. Aquaculture 415:183-190.
Ngugi CC, Fitzsimmons K, Manyala J, Bundi JM, Kimotho AN, Amadiva JM, Ndogoni JN, Munguti J (2018). Assessment of growth performance of monosex Nile tilapia (*Oreochromis niloticus*) in cages using low-cost, locally produced supplemental feeds and training fish farmers on best management practices in Kenya. Project Report. Sustainable Feed Technology and Nutrient Input Systems/SFT/13SFT06AU.
Nyandat B, Owiti G (2013). Aquaculture needs assessment mission report. Report/Rapport: SF-FAO/2013/24. September/Septembre 2013. FAO-SmartFish Programme of the Indian Ocean Commission, Ebene, Mauritius.
Obiero KO, Waidbacher H, Nyawanda BO, Munguti JM, Manyala JO, Kaunda-Arara B (2019). Predicting uptake of aquaculture technologies among smallholder fish farmers in Kenya. Aquaculture International 27:1689-1707.
Obwanga B, Rurangwa E, Soma K, Kilelu C (2018). A comparative study of aquaculture sector development in Egypt, Ghana and Nigeria: Insights for Kenya's sustainable domestic sector development. Wageningen University and Research. Research Report 006.
Obwanga B, Lewo MR (2017). From aid to responsible trade: driving competitive aquaculture sector development in Kenya; Quick scan of robustness, reliability and resilience of the aquaculture sector. Wageningen University and Research, Report 2017-092 3R Kenya
Okechi JK (2004). Profitability assessment: A case study of African catfish (*Clarias gariepinus*) farming in the Lake Victoria Basin, Kenya. (Master's thesis, The United Nations Univerist
Ole-Moiyoi LK (2017). Fishing for answers: can aquaculture transform food security in rural Kenya? PhD Dissertation. Standford University, Stanford, USA. P. 163.
Olufayo M (2012). The gender roles of women in aquaculture and food security in Nigeria. Visible possibilities: The economics of sustainable fisheries, aquaculture and seafood trade: Proceedings of the Sixteenth Biennial Conference of the International Institute of Fisheries Economics and Trade, July 16-20, Dar es Salaam, Tanzania. Edited by Ann L. Shriver. International Institute of Fisheries Economics and Trade (IIFET), Corvallis, USA.
Peterman A, Behrman J, Quisumbing A (2010). A review of empirical evidence on gender differences in non-land agricultural inputs, technology, and services in developing countries', IFPRI Discussion Paper 975. Washington DC: International Food Policy Research Institute. https://doi: 10.1007/978-94-017-8616-4_7
Phillips M, Subasinghe RP, Tran N, Kassam L, Chan CY (2016). Aquaculture big numbers. FAO Fisheries and Aquaculture Technical Paper No. 601. FAO, Rome, Italy. http://www.fao.org/3/a-i6317e.pdf.
Rubin D, Manfre C, Barrett K (2009). Promoting gender equitable opportunities in agricultural value chains. Handbook, Washington, DC, USA.
Rutaisire J, Kabonesa C, Okechi JK, Boera PN (2010). Lake Victoria Basin: Gender issues in fish farming. In: Flintan, F., Tedla, S. (Eds.), Natural Resthece Management. The impact of Gender and Social Issues. Published by the International Development Research Centre (IDRC).
Schumacher KP (2014). Gender relations in global agri-food value chains–A review. Erde. https://doi.org/10.12854/erde-145-10.
SDF, State Department of Fisheries (2012). Fisheries Annual Statistical Bulletin 2012. Ministry of Agriculture, Livestock and Fisheries, Nairobi, Kenya.
Sexsmith KSC, Speller W (2017). How to improve gender equality in agriculture, Investment in agriculture policy brief No 5. International Institute for Sustainable Development.
van Eerdewijk A, Wong F, Vaast C, Newton J, Tyszler M, Pennington A (2017). White Paper: A conceptual model of women and girls' empowerment. Technical Report. March, 2017. https://www.researchgate.net/publication/315516870
Veliu A, Gessese N, Ragasa C, Okali C (2009). Gender analysis of aquaculture value chain in Northeast Vietnam and Nigeria. Agriculture and Rural Development, Discussion Paper 44. The World Bank, Washington. http://siteresources.worldbank.org/INTARD/Resources/Gender_Aquaculture_web.pdf.
UNDP, United Nations Development Program (2018). Human development indices and indicators: 2018 statistical update. New York.

Weeratunge N, Chiuta T, Choudhury A (2012). Transforming aquatic agricultural systems (AAS) towards gender equality: A five country review. Working Paper, CRP1.3 Aquatic Agricultural Systems, The WorldFish Center, Penang. P. 83.

Weeratunge-Starkloff N, Pant J (2011). Gender and aquaculture: Sharing the benefits equitably. The WorldFish Center, Penang, Malaysia. Issues Brief, 2011-32. P. 12.

Weeratunge N, Snyder KA, Choo PS (2010). Gleaner, fisher, trader, processor: understanding gendered employment in fisheries and aquaculture. Fish and Fisheries 11:405-420.

Weeratunge N, Snyder KA (2009). Gleaner, fisher, trader, processor: Understanding gendered employment in the fisheries and aquaculture sector. Paper presented at the FAO-IFAD-ILO Workshop on "Gaps, trends and current research in gender dimensions of agricultural and rural employment: differentiated pathways out of poverty", Rome, 31st March-2nd April 2009.

Williams SB (2000). Economic potentials of women in small scale fisheries in West Africa. Obafemi Awolowo University, Nigeria. IIFET 2000 Proceedings.

World Bank (2012). World Development Report, 2012: Gender equality and development. World Bank; Washington, DC: https://openknowledge.worldbank.org/handle/10986/4391. [Google Scholar]

Yin RK (2009). Case study research: Design and methods (4th Ed.). Thousand Oaks, CA: Sage. https://doi.org/10.33524/cjar.v14i1.73

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Inoculant production of *Pisolithus* sp. in submerged culture under agitation

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Ectomycorrhizal fungi inoculation can increase the sustainability of planted forests hence the inoculant production should be optimized. The objective of this study was to determine the agitation speed and time of growth for maximum production of mycelium of D216 isolate of *Pisolithus* sp. The treatments were established by factorial 6x3, being six agitation conditions and three different times (days) of growth. The kinds of agitation used were: Without agitation, gentle manual agitation every two days and orbital agitation speed of 50, 100, 200 and 250 rpm. The times of growth were 14, 21 and 28 days. The mycelium mass was generally single and remained on the surface of the culture medium when grown without agitation or with manual agitation. Spherical mass units formed under agitation and grew submerged. The dry mass production of mycelium of D216 isolate was higher when grown under agitation of 200 rpm, followed by 250 and 100 rpm. For all agitation system, the production of mycelium did not increase after 14 days. The maximum production of mycelium dry mass of D216 isolate of *Pisolithus* sp. was obtained in the agitation speed of 200 rpm for 14 days. The elevation of the agitation to 250 rpm decreased the mycelium dry mass production of *Pisolithus* sp. In the absence or in lower agitation speeds, the mycelium dry mass was minimal. Using the ideal speed for maximum mycelium production and adjusting other growing conditions can decrease the risk of contamination, the production time and cost of the inoculant and encourage the use of this biotechnology.

Key words: Ectomycorrhizal fungi, inoculation, mycorrhiza, planted forests.

INTRODUCTION

The ectomycorrhizal fungi (EMF) are well known due to the association ability with planted trees species, such as

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eucalyptus and pines, increasing the survival and growth of the same (Chen et al., 2006; Quoreshi et al., 2008; Rivero et al., 2009; Costa et al., 2015, 2019). The inoculation of these trees with selected ectomycorrhizal fungi is a biotech strategy that can increase productivity in the forestry sector and reduce the excessive use of chemical fertilizers, which contribute to the high production cost and environmental impacts (Alberton et al., 2014; Costa et al., 2019). In ectomycorrhizae, fungi increase the plants ability to absorb water and nutrients and to withstand biotic and abiotic stresses and plants provides carbohydrates to the fungi (Garbaye, 2000; Chalot et al., 2002).

Inoculation of selected ectomycorrhizal fungi can reduce the use of fertilizers by up to 33% and increase coniferous seedlings growth, thus contributing to the reduction of environmental impacts associated with the production and use of chemical fertilizers in nurseries (Khasa et al., 2001). The beneficial effect of inoculation of ectomycorrhizal fungi in nursery was also observed in *Eucalyptus* seedlings (Chen et al., 2006; Costa et al., 2019), as well as the improvement in the quality and performance of transplanted seedlings (Quoreshi et al., 2008; Rivero et al., 2009). Inoculation of *Pisolithus microcarpus* isolate UFSC-Pt116 in *Eucalyptus* cutting increased plant growth by 13.1% at commercial nursery (Costa et al., 2019). Furthermore, seedlings of *Pinus radiata* inoculated with *Rhizopogon roseolus* and *Scleroderma citrinum* (Ortega et al., 2004) and seedlings of *Pinus pinea* inoculated with *Rhizopogon luteolus* and *Rhizopogon roseolus* (Parladé et al., 2004) survived and grew more than those not inoculated after planting in the field. Despite these results, the lack of investment by both government agencies and companies in the forestry sector limits the production and use of ectomycorrhizal inoculants in commercial nurseries in Brazil (Rossi et al., 2007).

The mycelium is the most appropriate way for these fungi inoculation (Brundrett et al., 1996), hence it must be used directly in *in vitro* tests, in selection tests in a greenhouse and commercially in the nursey seedlings inoculation (Alves et al., 2001). Thus, for its large-scale use, it is necessary to produce mycelium from pure cultures in culture medium (Pokojska et al., 1996).

Based on the assumption that the ectomycorrhizal fungus requires contact with nutrients and oxygen to grow, and considering the possibility of biotechnological application of biomass produced and the various uses of mycelium, the cultivation in a liquid culture medium is the most suitable for the mycelium biomass production (Rosado et al., 2003; Repáč, 2011). This cultivation can also be done in less space and less time when compared with the solid medium (Martin, 1983; Papagianni, 2004; Albaek et al., 2011). In addition, it offers ease of mycelium separation from the culture medium, allows the growth medium sterilization, maintenance of the aseptic culture and greater cultivation conditions control during the production process (Guillén-Navarro et al., 1998).

The physical effects of shaking speed during the fungi cultivation in a liquid culture medium require a great deal of attention. There are several reports that the agitation speed can change the morphology, differentiation of filamentous species and the time of growth and mycelium production of the fungi (Park et al., 2002b; Kim et al., 2003; Kelly et al., 2004; El-Enshasy et al., 2006). The agitation is important for the homogenization of liquid culture medium, mixing of nutrients, mass transfer and heat between the different phases present in the culture and maintenance of physical and chemical conditions suitable for mycelium production (Mantzouridou et al., 2002; Gupta et al., 2003). Furthermore, it is directly linked to the aeration rate of the culture medium, which may be beneficial for the growth and performance of the cells (Park et al., 2002a; Kim et al., 2003).

On the other hand, the exposure of the mycelium to high agitation speeds create shear forces, which undermine the fungi in several ways, reducing the growth and, so, the mycelium biomass production (Park et al., 2002b; Gupta et al., 2003; Kelly et al., 2004; El-Enshasy et al., 2006). However, there are also reports that the increase of agitation speed increases mycelium production (Cui et al., 1997; Kim et al., 2003). Although the fungi responses to unrest have been quite varied, the general trend for both the agitation and aeration is a positive correlation with the mycelium production (Albaek et al., 2011).

The use of the ideal agitation speed for the growth of each fungus isolate can increase mycelium production in less time and thus reduce production costs (Amanullah et al., 2000; Albaek et al., 2011). Among the ectomycorrhizal fungi, the species of the *Pisolithus* are among the most studied regarding the mycorrhizae formation (Barros et al., 1978; Campos et al., 2011; Alberton et al., 2014). The influence of agitation speed at the growth rate and mycelium formation should be examined, with the purpose of finding which agitation speed results in increased mycelium production in less time, causing less damage to the fungal cells. In this context, the objective of this study was to determine the agitation speed and the time of growth for maximum production of mycelium of D216 isolate of *Pisolithus* sp. in liquid culture medium.

MATERIALS AND METHODS

Location of experiment and fungal isolate

The study was carried out in the Laboratory of Soil Microbiology from the Federal University of Jequitinhonha and Mucuri Valleys - UFVJM, in Diamantina, Minas Gerais, Brazil (with headquarters at 18.20°S and 43.57°W).

The D216 isolate of the *Pisolithus* sp. used isolate that was obtained from EMF collection of the Laboratory of Soil Microbiology – UFVJM. Pure isolate cultures were originally obtained from basidiomas sampled in *Eucalyptus* spp. plantations in

Jequitinhonha Valley – Minas Gerais. It was selected by presenting, in previous experiments, satisfactory growth in solid and liquid culture medium and by promoting benefits to eucalyptus plants *in vitro* tests (Costa et al., 2015) and in the nursery (Gomes, 2016).

Isolate growth, experimental design and treatments

The isolate was grown in Petri dishes (∅= 100 mm) with 20 mL of solid MNM culture medium for 29 days at 25ºC. Subsequently, discs of 5 mm diameter were removed from the edges of the isolate colonies, transferred to Petri dishes and kept for three more days to allow the damaged mycelium recovery during transplanting, confirm the feasibility, demonstrate the absence of contamination and perform the pre-growth for cultivation in liquid medium.

The experiment was carried out in a completely randomized design and treatments were established by factorial 6x3, being six agitation conditions and three different times (days) of growth. The agitations used were: without agitation, gentle manual agitation for three seconds every two days and orbital agitation speed of 50, 100, 200 and 250 rpm. The times of growth were 14, 21 and 28 days. The experimental unit was composed of one Erlenmeyer containing 50 mL of liquid Melin-Norkrans modified culture medium (MNM), pH 6,5 (Marx, 1969) inoculated with 10 discs of 5-mm diameter culture medium with pre-grown mycelium of D216 isolate of *Pisolithus* sp. and incubated at 25°C. Each treatment had eight repetitions, totaling 144 experimental units. The MNM culture medium was added of 20 mg L^{-1} of chloramphenicol to prevent bacterial contamination.

Determination of the mycelium dry mass and data analysis

To each time of cultivation, the fungal mycelium originated from each Erlenmeyer was collected in a sieve with a mesh of 53 µm, washed with distilled water. At the time of washing, the mycelium was visually analyzed for density, branching and fragmentation. The material was transferred to 30 mL plastic containers previously weighed and dried in an oven at 60ºC for three days until constant weight (Brundrett et al., 1986). Then, the set was weighed on an analytical balance (Marte, AY220). The mycelium dry mass was determined by the difference between the final weight of the plastic container with dried mycelium and the initial weight of the empty plastic container and was expressed in milligrams by 50 mL of culture medium (experimental unit).

Dried mass of mycelium was analyzed for distribution (Lilliefors test) and homogeneity (Cochran and Bartlett test). Then, using the Sisvar software, dates were subjected to two-way ANOVA and the means compared by the Tukey test (5% significance).

RESULTS

The agitation conditions influenced the mycelium dry mass production of D216 isolate of *Pisolithus* sp., however, the agitation with time interaction was not significant (Table 1).

The mycelia morphology, mycelium density or branching frequency, of the D216 isolate was visually different in the agitation condition evaluated. When grown without agitation or manual agitation some of the mycelium discs placed in the medium did not grow, the mycelium mass (densely interwoven mycelium masses referred to here as pellet) was generally single and remained on the surface of the culture medium. Differently, under agitation, several spherical pellets units were formed and these grew submerged. At the end of the cultivation, these pellets differed in size and mycelium density; at a speed of 50 rpm, they were smaller and some fragmented hyphae filaments also occurred dispersed. At the speed of 100 rpm, mycelia pellets were larger than the previous ones and they had a hairy hyphal layer. At the speed of 200 rpm, the mycelia pellets were larger than the previous ones and had a more mycelium density with a more branching frequency. At the speed of 250 rpm, the mycelia pellets were smaller, irregular, and there were more hyphae fragmented.

The dry mass production of mycelium of D216 isolate was higher when grown under agitation of 200 rpm (747 mg), followed by 250 rpm (510 mg) and 100 rpm (157 mg) (Figure 1). For all agitation conditions, the production of mycelium did not increase after 14 days. The maximum production of mycelium dry mass (757 mg) of D216 isolate of *Pisolithus* sp. was obtained in the agitation speed of 200 rpm at 14 days of growth, while in the absence or low agitation speed the mycelium mass production was not affected (Figure 1).

Elevation of the agitation speed from 200 to 250 rpm resulted in a decrease in the mycelium production of *Pisolithus* sp. (Figure 1).

DISCUSSION

The influence of agitation speed on the morphology of filamentous fungi growing in submerged liquid culture medium was also observed for *Blakeslea trispora* (Mantzouridou et al., 2002), *Cordyceps militaris* (Park et al., 2002b), *Paecilomyces sinclairii* (Kim et al., 2003) and *Aspergillus niger* (Kelly et al., 2004). This effect of the agitation speed on the fungi morphology confirms that these are morphologically complex organisms, differing in structure at different times in their life cycle, differing in form between surface and submerged growth, differing also with the nature of the growth medium and physical environment (Papagianni, 2004). Filamentous fungi growing in low agitation speeds, in general, produce larger pellets and lower mycelium dry mass, probably due to the lower nutrient availability in the internal parts of the pellet (El-Enshasy et al., 2006).

Regardless of the agitation conditions, there was dry mass production of the D216 isolate of *Pisolithus* sp. until the 14 days of growth (Figure 1). The insignificant growth of mycelium after 14 days of cultivation may be due to changes in the pH of the culture medium or depletion of some nutrient. This higher growth rate until the 14 days is better than what is cited in the literature for *Pisolithus* sp., which in general,is grown between 20 and 30 days for the inoculant production (Fernandes et al., 2014; Costa et al., 2015). For other faster growth fungi, as *A. niger* and *Ceriporiopsis subvermispora*, the greater quantity of mycelium dry mass is produced in a shorter time, from four to five days (Gupta et al., 2003; Kelly et al., 2004).

Table 1. Analysis of variance and respective levels of significance for the dry mass of mycelium of the isolate D216 of *Pisolithus* sp. cultured in modified Melin-Norkrans liquid medium under gentle manual shaking every two days and orbital shaking of 0 (without stirring) 50, 100, 200 and 250 rpm and growth times of 14, 21 and 28 days.

Variation source	DF	Mean square	F	P value
Agitation conditions	5	2246689.51*	700.29	0.0000
Growth time	2	3309.31	1.032	0.3595
Agitation conditions x Growth time	10	525.35	0.164	0.9983
Error	126	3208.23		
Average mass of dry mycelium(mg in 50 mL)		256.17		
CV (%)		22.1		

*Significant at 5% by the Tukey test.

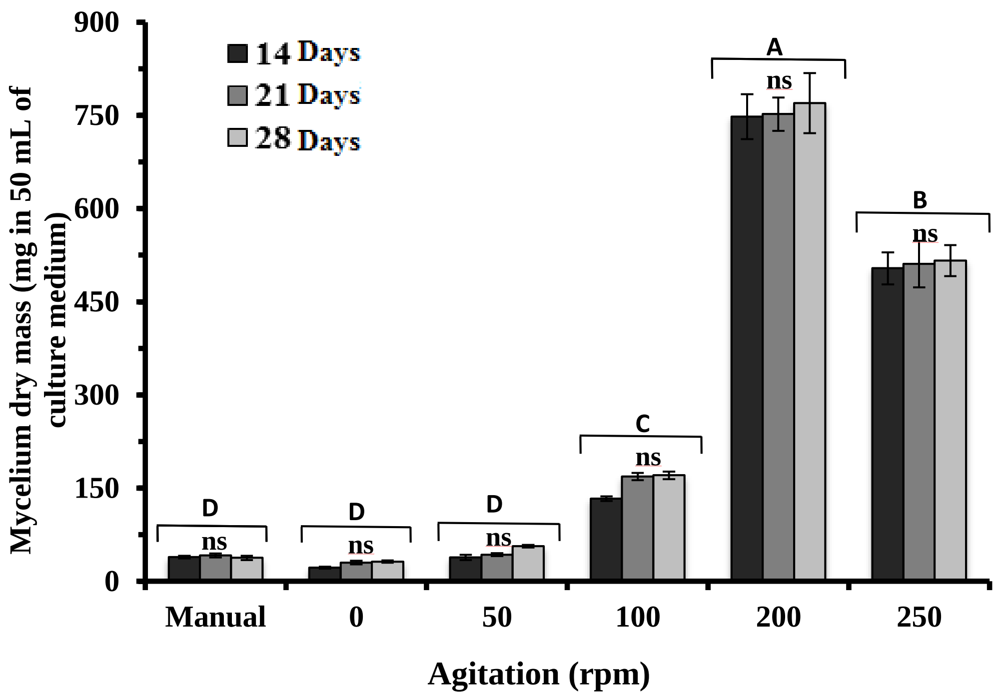

Figure 1. Dry mass of mycelium of the isolate D216 of *Pisolithus* sp. cultured in 50 mL of modified Melin-Norkrans liquid medium under manual shaking every two days and orbital shaking of 0 (without stirring) 50, 100, 200 and 250 rpm and growth times of 14, 21 and 28 days. ns = Growth times within each shaking system were not significant by the Tukey test ($p \leq 0.05$). The agitation systems followed by the same letter do not differ by the Tukey test ($p \leq 0.05$). The bars indicate standard error ($n = 8$).

Those authors proposed that the reduction in the growth rate was due to the depletion of the glucose.

The growth of the D216 isolate of *Pisolithus* sp. was minimal when incubated without agitation, agitated manually every two days or under agitation speed of 50 rpm even grown for 28 days (Figure 1). At a speed of 50 rpm, they were smaller and some fragmented hyphae filaments also occurred dispersed. This means that the same requires agitation for its greater growth in liquid medium. The lack of homogeneity of the liquid culture medium may have interfered with the aeration and distribution rate of nutrients, hindering the mycelium growth and consequently the dry mass production of *Pisolithus* sp. The agitation speed influences the nutrient availability and aeration rates, which reflects, in the mycelium morphology and increases the hyphae length, and consequently also the mycelium dry mass production (Amanullah et al., 1999, 2000; Park et al., 2002a, b; Mantzouridou et al., 2002; Gupta et al., 2003; Kelly et al., 2004; El-Enshasy, 2006). The agitation results in better culture medium mixing and maintains the concentration gradient between the cells' interior and exterior. This gradient promotes the satisfactory supply of sugars and other nutrients to the cells, besides facilitating the removal of gases and other catabolism byproducts from the cellular microenvironment, resulting in greater mycelium dry mass production (Mantzouridou et al., 2002).

The agitation speed increase for 100 and 200 rpm increased the visible number of mycelium pellets and mycelium dry mass of *Pisolithus* sp., reaching maximum production at a speed of 200 rpm (Figure 1). At the speed of 200 rpm, the mycelia pellets were larger than the previous ones and had a more mycelium density and seemingly more branching frequency. The 747 mg of dry mycelium mass obtained in this work at a speed of 200 rpm was 3.9 times greater than that observed for *Laccaria laccata* grown in 100 mL of liquid Lamb's medium inoculated with one 10-mm in diameter disc without agitation (Pokojska et al., 1996) and was 21 times greater than that observed for the ectomycorrhizal fungi *Cadophora finlandia* in solid MNM modified (Azaiez et al., 2018). This effect of increased agitation speed was also observed for the pathogenic fungus *B. trispora* (Mantzouridou et al., 2002) and the parasitic fungus *C. militaris* (Park et al., 2002b). The authors' proposed that increase of mycelium dry mass was due to better air supply to the cells. Generally, aerobic fungi have an increased growth rate when moderate aeration is combined with an increase in the agitation speed (Mantzouridou et al., 2002).

With the additional increase of the agitation speed to 250 rpm, the mycelia pellets were smaller, irregular, and there were more hyphae fragments and decreased mycelium dry mass production (Figure 1). Although, for other fungi, *Aspergillus awamori* (Cui et al., 1997) and *P. sinclairii* (Kim et al., 2003), the agitation speed of 250 rpm increased the mycelium dry mass production. The increase in the number of pellets under higher agitation speeds was also observed in *A. niger* growing at speeds of 200 to 800 rpm (El-Enshasy et al., 2006). Most of the time, agitation speeds lead to greater energy dissipation, linked to high shear risk, which can result in cell fragmentation and damage, decreasing the mycelium production (Park et al., 2002b; Gupta et al., 2003; Kelly et al., 2004; El-Enshasy et al., 2006). In addition, it can increase the culture medium's apparent viscosity, due to the increase in the quantities of free filaments, causing the reduction of the efficiency in mass transfer and limitingoxygen and nutrient absorption by the cells (Kelly et al., 2004).

The results obtained in this study confirmed that agitation speed is an important parameter in the mycelium development and dry mass production of *Pisolithus* sp., and indicates the agitation speed of 200 rpm as the ideal for growth and production of mycelium dry mass of 216 isolate of *Pisolithus* sp. These results corroborate with those found in literature, where it is reported that in appropriate aeration conditions, the optimal agitation speed for mycelium mass production in general is within the range of 150 to 250 rpm (Papagianni et al., 2001; Mantzouridou et al., 2002; Park et al., 2002a; Kim et al., 2003; El-Enshasy et al., 2006). The agitation speed within this range provides a balance between the forces of cohesion and disintegration; besides allowing the formation of small pellets favorable to a higher mycelium dry mass production during prolonged cultivation (Papagianni et al., 2004; El-Enshasy et al., 2006).

Conclusion

The mycelium dry mass production of D216 isolate of *Pisolithus* sp. is maximum at 14 days of growth under 200 rpm of agitation. Using the ideal speed for maximum mycelium production and adjusting other growing conditions can decrease the risk of contamination, the production time and cost of the inoculant and encourage the use of this biotechnology.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Albaek MO, Gernaey KV, Hansen MS, Stocks MS (2011). Modeling enzyme production with *Aspergillus oryzae* in pilot scale vessels with different agitation, aeration, and agitator types. Biotechnology Bioengineering 108(8):1828-1840.

Alberton O, Aguiar D, Gimenes MRT, Carrenho R (2014). Meta-analysis for responses of eucalyptus and pine inoculated with ectomycorrhizal fungi in Brazil. Food Agriculture Environment Journal 12(2):1159-1163.

Alves JR, Souza O, Podlech PAS, Giachini AJ, Oliveira VL (2001). Efeito de inoculante ectomicorrízico produzido por fermentação semi-sólida no crescimento de *Eucalyptus dunnii* Maiden. Pesquisa Agropecuária Brasileira 39(4):307-313.

Amanullah A, Blair R, Nienow AW, Thomas CR (1999). Effects of agitation intensity on mycelial morphology and protein production in chemostat cultures of recombinant *Aspergillus oryzae*. Biotechnology Bioengineering 62(4):434-446.

Amanullah A, Jüsten P, Davies A, Paul GC, Nienow AW, Thomas CR (2000). Agitation induced mycelial fragmentation of *Aspergillus oryzae* and *Penicillium chrysogenum.* Biotechnology Bioengineering 5(2):109-114.

Azaiez A, Nadeau MB, Bertrand A, Khasa DP (2018). In vitro selection of ecologically adapted ectomycorrhizal fungi through production of fungal biomass and metabolites for use in reclamation of biotite mine tailings. Mycologia 110(6):1017-1032.

Barros NF, Brandi RM, REIS MS (1978). Micorriza em eucalipto. Uma revisão sobre a morfologia, a fisiologia e os efeitos mútuos da associação fungo-planta. Revista Árvore 2(2):130-140.

Brundrett M, Bougher N, Dell B, Grove T, Malajczuk, N (1996). Working with Mycorrhizas in Forestry and Agriculture. Canberra: ACIAR Monograph 32 - Australian Centre for International Agricultural Research 374 p.

Campos DTS, Silva MCS, Luz JMR, Telesfora RJ, Maria CMK (2011). Colonização micorrízica em plantios de eucalipto. Revista Árvore 35(5):965-974.

Chalot M, Javelle A, Blaudez D, Lambilliote R, Cooke R, Sentenac H, Wipf D, Botton B (2002). An update on nutrient transport processes in ectomycorrhizas. Plant and Soil 244(2):165-175.

Chen YL, Kang LH, Malajczuk N, Dell B (2006). Selecting ectomycorrhizal fungi for inoculating plantations in South China: effect of *Scleroderma* on colonization and growth of exotic *Eucalyptus globulus, E. urophylla, Pinuselliottii*, and *P. radiata*. Mycorrhiza. New Phytologist 16(4):251-259.

Costa LS, Grazziotti PH, Grazziotti DCFS, Silva AC, Rossi MJ, Silva EB, Costa VHD, Gomes ALF (2015). *In vitro* evaluation of *Eucalyptus* ectomycorrhizae on substrate with phosphorus doses for fungal pre-selection. Revista Árvore 39(1):127-136.

Costa LS, Grazziotti PH, Silva AC, Fonseca AJ, Gomes ALF, Grazziotti DCFS, Rossi MJ (2019). Alginate gel entrapped ectomycorrhizal inoculum promoted growth of cuttings of *Eucalyptus* clones under nursery conditions. Canadian Journal of Forest Research 48(8):978-985.

Cui YQ, Van der Lans RGJM, Luyben KCAM (1997). Effect of agitation intensities on fungal morphology of submerged fermentation. Biotechnology Bioengineering 55(5):15-725.

El-Enshasy H, Kleine J, Rinas U (2006). Agitation effects on morphology and protein productive fractions of filamentous and pelleted growth forms of recombinant *Aspergillus niger*. Process Biochemistry 41(10):2103-2112.

Fernandes MCS, Costa LS, Grazziotti PH, Grazziotti DCFS, Santos JB, Rossi MJ (2014). *Pisolithus* sp. tolerance to glyphosate and isoxaflutole *in vitro*. Revista Árvore 38 (3):461-468.

Garbaye J (2000). The role of ectomycorrhizal symbiosis in the resistance of forest to water stress. Outlook on Agriculture 29(1):63-69.

Guillén-Navarro GK, Márquez-Rocha FJ, Sanchez-Vázquez JE (1998). Producción de biomasa y enzimas ligninolíticas por Pleurotus ostreatus en cultivo sumergido. Revista Ibero americana de Micología 15:302-306.

Gupta R, Gigras P, Mohapatra H, Goswami VK, Chauhan B (2003). Microbialα-amylases: biotechnological perspective. Process Biochemistry 38(11):1599-1616.

Kelly S, Grimm LH, Hengstler J, Schultheis E, Krull R, Hempel DC (2004). Agitation effects on submerged growth and product formation of Aspergillus niger. Bioprocess and Biosystems Engineering 26:315-323.

Khasa PD, Sigle L, Chakravarty P, Dancik BP, Erickson L, Mc Curdy D (2001). Effect of fertilization on growth and ectomycorrhizal development of container-grown and bare-root nursery conifer seedlings. New Forests 22:179-197.

Kim SW, Hwang HJ, Xu CP, Choi JW, Yun JW (2003). Effect of aeration and agitation on the production of mycelial biomass and exopolysaccharides in an enthomopathogenic fungus *Paecilomyces sinclairii*. Letters Applied Microbiology 36(5):321-326.

Mantzouridou F, Roukas T, Kotzekidou P (2002). Effect of the aeration rate and agitation speedon β-carotene production and morphology of *Blakesleatrispora*in a stirred tank reactor: mathematical modeling. Biochemical Engineering Journal 10(2):123-135.

Martin AM (1983). Submerged production of edible mushroom mycelium. Canadian Institute Food Science Technology Journal 16(3):215-217.

Marx DH (1969). The influence of ectotrophicmycorrhyzal fungi on the resistence of pine roost to pathogenic infections and antagonism of mycorrhizal to root pathogenic fungi and soil bacteria. Phytopathology 59(2):153-163.

Ortega U, Duñabeitia M, Menendez S, Gonzalez-Murua C, Majada J (2004). Effectiveness of mycorrhizal inoculation in the nursery on growth and water relation of *Pinus radiata* in different water regimes. Tree Physiology 24(1):65-73.

Papagianni M, Nokes SE, Filer K (2001). Submerged and solid-state phytase fermentation by *Aspergillus niger*: Effects of agitation and medium viscosity on phytase production, fungal morphology and inoculum performance. Food Technolology and Biotechnology 39(4):319- 326.

Papagianni M (2004). Fungal morphology and metabolite production in submerged mycelial processes. Biotechnology Advances 22(3):189-259.

Park JP, Kim YM, Kim SW, Hwang HJ, Cho YJ, Lee YS, Song CH, Yun JW (2002a). Effect of aeration rate on the mycelial morphology and exo-biopolymer production in *Cordyceps militaris*. Process Biochemistry 37:1257-1262.

Park JP, Kim YM, Kim SW, Hwang HJ, Cho YJ, Lee YS, Song CH, Yun JW (2002b). Effect of agitation intensity on the exo-biopolymer production and mycelial morphology in *Cordyceps militaris*. Letters Applied Microbiology 34(6):433-438.

Parladé J, Luque J, Pera J, Ricón AM (2004). Field performance of *Pinuspinea* and *P. halepensis* seedlings inoculated with *Rhizopogon* spp. and outplanted in formerly arable land. Annals Forest Science 61(6):507-514.

Pokojska A, Kampert M, Różycki H, Strzelczyk E (1996). Effects of vitamins, temperature and pH on the biomass production by ectomycorrhizal fungi. Acta Mycologica 31(1):55-65.

Quoreshi AM, Piché Y, Khasa DP (2008). Field performance of conifer and hardwood species 5 years after nursery inoculation in the Canadian Prairie Provinces. New Forest 35(3):235-253.

Repáč I (2011). Ectomycorrhizal inoculum and inoculation techniques. In. Rai M, Varma A (Eds.), Diversity and Biotechnology of Ectomycorrhizae: Soil Biology, pp. 43-63.

Rivero SHT, Moorillón VGN, Borunda EO (2009). Growth, yield, and nutrient status of pecans fertilized with biosolids and inoculated with rizosphere fungi. Bioresource Technology 100(6):1992-1998.

Rosado FR, Germano S, Carbonero ER, Costa SMG, Iacomini M, Kemmelmeier CD (2003). Biomass and exopolysaccharide production in submerged cultures of *Pleurotus ostreatoroseus* Sing. and *Pleurotus ostreatus* "florida" (Jack.: Fr.) Kummer. Journal Basic Microbiology 43(3):230-237.

Rossi MJ, FurigoJr A, Oliveira VL (2007). Inoculant production of ectomycorrhizal fungi by solid and submerged fermentations. Food Technology Biotechnology 45(3):277-286.

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