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Review

Irrigation potentials and rice self-sufficiency in Nigeria: A review

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Nigeria has abundant land and water resources to embark on irrigation schemes to ensure all year round rice production. Additional yield to annual rice production through dry season farming in 2012 in 10 pilot States of the country was found to be over 1 million metric tonnes. This shows that an intensive and consistent irrigated rice production scheme will set Nigeria on a pathway to rice self-sufficiency thereby bringing an end to the gross loss in foreign exchange due to importation and smuggling of the commodity. The study reviewed irrigation subsector in Nigeria, and revealed that the prospects of achieving rice self sufficiency through irrigation farming is hampered by some major challenges namely-underdeveloped subsector promoted by fragmented, inconsistent and unimplemented policies, multiple water regulatory institutions with overlapping and duplicating mandate and poor management system. Other challenges include- the absence of a viable market for local rice as well as high cost of labour inputs, irrigation equipments and other operating costs. The study concluded that for Nigeria irrigation potentials to be harnessed towards rice self-sufficiency there is need to amend policies on irrigation and water resources, create conducive market for local producers and provide subsidized and appropriate farm implements.

Key words: Irrigation potentials, rice self-sufficiency, challenges, Nigeria.

INTRODUCTION

Nigeria with an estimated 183, 523, 43 people is the most populous country in Africa (World Bank, 2014a). Increase in population growth rate, youth bulge, increasing urbanization, changes in dietary needs of the people as well as being a staple food has resulted in increase in the consumption of rice.

As a staple food in the diet of most Nigerians, there is a high demand for rice which has cumulated in a huge gap between the supply and demand for rice. In the last decade rice consumption has increased at an annual average rate of 10.3% (Maji et al., 2015). The per capita consumption of rice has grown from 3 kg in the 1960s to an estimated 37.5 kg in 2014 (UARK, 2015) and is expected to increase due to increase in population and urbanization.

Though production has also increased over the years from an average of 300,000 tonnes in the 1990s to over 4 million tonnes in the year 2013 (FAOSTAT, 2015) the

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increase in production has being by expansion in area harvested to rice which has increased from 14,000ha in the 1960s, and has grown through the years to 2,863.815 ha in the year 2013. But to meet the demand of the growing population, Talpur (2011) is of the view that intensification of yield from each unit of land harvested to a crop must be increased.

The inability of local supply to meet up with rice consumption needs has resulted in high imports of rice. The phenomenal rise in imports of 300 thousand tones annually in recent times has continued to drain the country of about ₦300 million annually in foreign reserves- on the average. Aside from the huge finances, rice imports exposes the country to international market shocks (in times of scarcity and rise in prices) thus with serious risk to food security.

To reduce the volume of imports and reduce external shocks, the Nigerian government over the years has formulated and used various policy instruments and interventions to boost local production. Some of these measures include: import restrictions, tariff restriction, and inauguration of presidential taskforce on rice (in 1980), inputs subsidies and ban on imports 1986-1995. These policies were put in place to stimulate local production and make local rice more competitive.

In 2003 the Federal Government set up a presidential initiative on rice production with the aim to become rice sufficient in 2007. The objective was to eliminate imports and generate exportable surplus and enhance food security through the production of 6 million tonnes of milled rice by the year 2005. A tariff of 100% was imposed then on rice imports (Daramola, 2005) and a levy of 10%.

Efforts to make the country become rice self sufficient was again renewed in 2010 which led to the formal lunch of the rice transformation strategy under the agricultural transformation agenda (ATA). The strategy was to produce more paddy and industrial grade milled rice that could compete with imported rice in the market. Since Nigeria has two production seasons; the raining season and the dry season. To this end, 268, 000 farmers were given leverage through subsidies in seeds, fertilizers, provision of watering pumps for irrigation farming in ten (10) states of the north namely: Niger, Kebbi, Sokoto, Kano, Zamfara, Bauchi Jigawa Katsina, kogi and Gombe. Other measures include; 35% tariff on brown rice imports, the establishment of fourteen integrated rice mills with capacity of 240,000 metric tonnes and also a tariff of 110% was imposed on rice imports in 2014. Also in June 23 2015 the CBN banned the sale of the foreign exchange to rice importers as a prohibition to discourage import.

Reports from the 2012 production activities in the 10 states showed that over 1 million metric tonnes of rice was harvested in the dry season irrigation farming (Adesina, 2013). A total of 2,170,000 metric tonnes was being expected from the dry and wet season farming from these 10 states. This has led to a downward review of 1.3 metric tonnes import quota for the year 2015 from 1.5 million metric tonnes of 2014. Though Nigeria has not attained self sufficiency as desired but a significant decrease has been observed in rice imports as shown in the proposed 2015 imports figures.

Most of the increase in local production has been attributed to irrigation farming embarked on in the 10 pilot states. Considering that Nigeria is well endowed with water and land resources for irrigation farming, utilization of these resources can close the demand supply gap of rice in the country. A considerable increase in production is essential for Nigeria to meet up with the growing demand considering its fast growing population. Meanwhile the Nigerian rice irrigation production has been left underdeveloped. Rice production generally in Nigeria is rain fed dependent, only about 293.000 ha of irrigable land has been equipped for irrigation and only about 218, 800 ha is being actually irrigated with about 173, 000 ha under private small scale while, 29,000 ha is under public irrigation scheme (FAO-Aquastat 2005).

An upgrade to irrigation will increase production significantly since it offers an opportunity for intensity in production of two to three times production in a year.

This paper therefore looks at challenges and also the potentials of irrigation farming in assisting Nigeria in attaining the desired rice self-sufficiency (Figures 1 and 2).

HISTORY OF IRRIGATION IN NIGERIA

The history of irrigated crop cultivation in Nigeria dates back to the Colonial era but became more pronounced after the drought of 1970-1975 (postcolonial era). The need for continuous cultivation to escape the hardship of food shortage and insecurity led to emphasis on irrigation practice. Irrigation practice was initially mostly traditional (used by small scale farmers) in form of FADAMA, gravity or natural flow, calabash/bucket and pump methods and the facilities were provided and maintained by farmers with no assistance from government or donor organizations (Yahaya, 2002; Umar, 1994). As the need for irrigated crop cultivation grew between 1972 and 1974, three pilot public irrigation schemes were developed namely; Bakolori scheme, Kano river irrigation scheme and the Chad Basin scheme (NINCID, 2015)

Consequently, the success of the pilot irrigation schemes in mitigating the adverse effect of the early 1970s drought further led to the insightful development of 12 River Basin Development Authorities (RBDAs) across the country by the Nigerian government (FAO and US Bureau, 1970). These RBDAs include the; Upper Benue Basin, Lake Chad Basin, Benin-Owena Basin, Sokoto-Rima Basin, Itadejia-Jema'a, Iyola Basin, Maiduguri Basin, Lower Benue Basin, Cross River Basin, Ogun-Osun Basin, Anambra-Imo, Niger Basin.

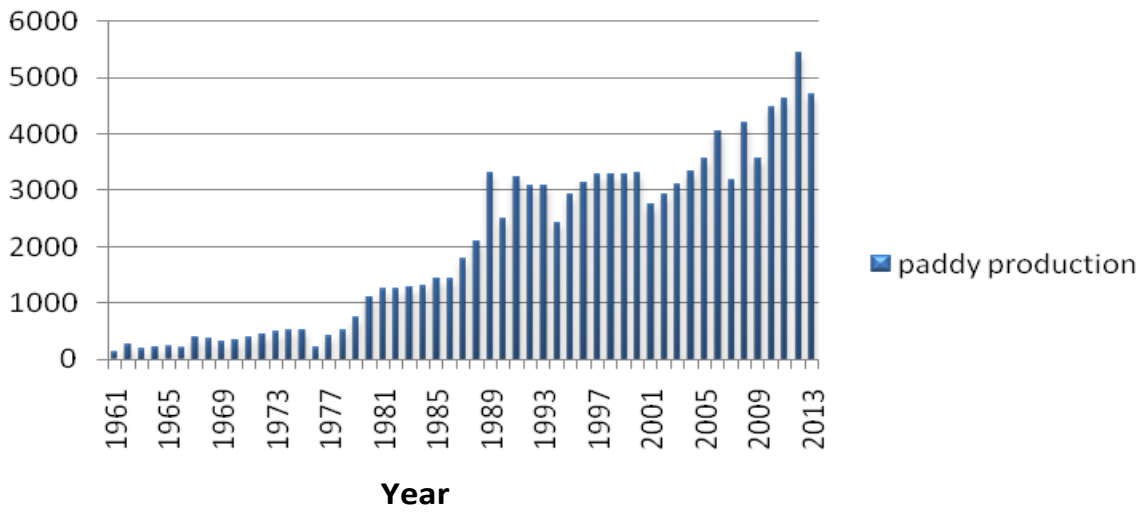

Figure 1. Rice paddy production in Nigeria 1961-2013. Authors illustration, data source: Fishfact (2014-2015).

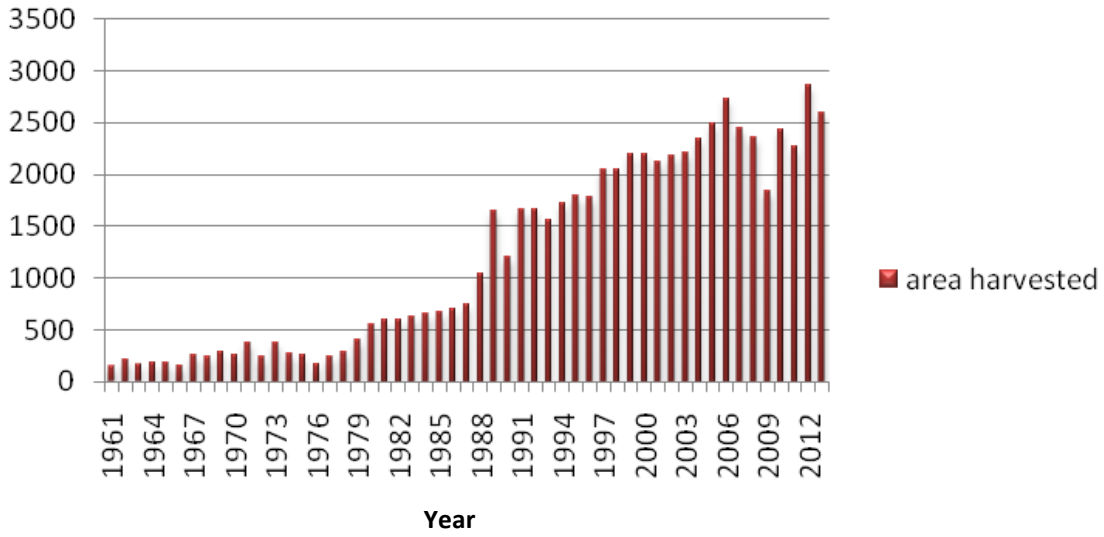

Figure 2. Area harvested to rice in Nigeria 1961-2013. Authors illustration, data source: Fishfact (2014-2015).

These RBDAs had the mandate of providing water for irrigation and domestic water supply, hydroelectric power generation, flood control, recreation facilities, fisheries projects and navigation improvement. RBDAs were also saddled with engendering big plantation farming and encouraging the establishment of industrial complexes that could bring about private-public business partnership, seed multiplication, livestock breeding and food processing. However, overburdening of the RBDAs with responsibilities and inconsistency in RBDAs policy framework has led to their inability to fulfill their core purpose of establishment as regards irrigated farming.

Until the establishment of these RBDAs, the water resource development for agricultural purposes was in the hands of the private sector. A notable example was in the production of sugar cane with few crops notably rice and vegetables by the States in Nigeria's northern part. Irrigation practice was mainly based on using residual flood waters and low lands (FADAMA-flash flood plains) and supplemented with the Shadoof (bucket-left system irrigation which originated from Sudan and introduced in Nigeria over a century ago). The major FADAMA areas were located along the flood plains of Rima, Sokoto, Niger, Benue and Yobe rivers.

Aside from the government, donor organisations like the World Bank and the Food and Agriculture Organisation (FAO) are today seen at the core of promoting irrigation practice in Nigeria. Promotion of

pumps and tube wells started in the late 1980s through the Agricultural Development Programmes (ADPs). By 1992 more than 80 thousand pumps each irrigating between 0.5-1ha had been distributed. From 1992 to 1999 the National FADAMA Development Project (NFDP) of the World Bank built on the ADP achievement and was able to distribute 55 thousand pump sets with an equipped area of 1ha per pump.

The FAO commenced a Special Food Security (SPFS) project in 1999 in three villages in Kano State through a participatory community development approach where farmers were responsible for planning and have ownership of the project. Motorized pumps and tube wells were provided for irrigation to meet the needs of farmers cultivating a total of 280 ha. After the success of this pilot scheme, the project was extended to the 36 States of the Federation and an additional 109 sites in 2002. By 2004, cultivated land area equipped for irrigation within the country was around 293117 ha (FAO, 2005).

Today, three main irrigation schemes exist in Nigeria they include: public irrigation schemes- which are systems controlled by the government and are popularly called formal irrigation, the farmer-owned and operated irrigation scheme, which receives assistance from government inform of subsidies and trainings (informal irrigation) and the residual flood plain FADAMA (purely traditional irrigation practice) which has now emerged in the World Bank assisted programme which started in the National FADAMA Development Programmes (NFDP).

While the initial purpose of irrigation development in Nigeria was based in part on the need to sustain growth in food supply that will in the long run lead to national food security, the attainment of this height is yet a mirage in rice production.

RICE PRODUCTION ECOLOGIES IN NIGERIA

Rice is produced in all agro ecological zones of Nigeria with the middle belt being the highest producer and enjoys comparative advantage in production over other parts of Nigeria (FAO, 2013). The potential land for rice production in Nigeria is between 4.6 to 4.9 million ha (Imolehin, 1991) of this rice is cultivated on an estimated 3.7 million ha covering about 10.6% of farmland that is under crop cultivation in the country (Cadoni et al., 2013). Rice production in Nigeria is majorly rain fed which represent about 77% of production. The rain fed production systems include:

Rain-fed upland ecology

Crops depend strictly on rain for growth and productivity in this ecology (Imolehin and Wada, 1999). This ecology accounts for about 30% of share of area cultivated to rice (Singh et al., 1997; Kebbeh et al., 2003), 17% share of domestic production and are characterized with average low yields of 1.7 tonnes/ha (Kebbeh et al., 2003).

Rain-fed low land ecology

Rice is produced in low land wet soils zone and it's the most favored ecology in the country given its resistance to drought (Kebbeh et al., 2003). An estimated 47% area is cultivated to lowland rice and it accounts for 57% of domestic production and an average yield of 2 tonnes/ha (Singh et al*.,* 1997; Kebbeh et al., 2003). Other rice producing ecologies include

Mangrove swamp

It covers 1% of land cultivated to rice but only 100ha has been developed so far (Imolehin and Wada, 1999). It contributes 1% to the total domestic production (Singh et al., 1997; Kebbeh et al., 2003) and characterized by low yields of about 2 tonnes/ha (Kebbeh et al., 2003).

Deep water floating rice ecology

This ecology covers about 5% of rice production area (Singh et al., 1997; Kebbeh et al., 2003), about 3% domestic share of rice and yields of about 1.3 tonnes/ha (Kebbeh et al., 2003).

Irrigated rice ecology

This ecology is the most recently developed ecology in Nigeria (Imolehin and Wada, 1999). This ecology is mostly found in the northern part of the country, irrigation water is being supplied from dams, bore holes and wells to supplement for rainfall which is not in much supply. This ecology accounts for about 17% of area share of rice production and 27% of domestic production. This ecology is characterized by average yields of 3.5 tonnes/ha but has high yields potentials of about 5-6 tonnes (Kebbeh et al., 2003) this ecology is noted to have best performance in terms of yields. It is therefore important that effort is made to develop this sector to its full potentials (Table 1).

IRRIGATION POTENTIALS IN NIGERIA

Water resources

About 221 km^3 of water resources are produced annually while total renewable water resources are estimated at 286.2 km^3 (FAO et al 2014). About 214 km^3 makes up surface water and the exploitable surface flow is about 96 km^3/ year while the volume of available groundwater

Table 1. Rice producing ecologies in Nigeria.

Production ecology	Major states covered	Estimated share of National rice-farmed area (%)	Share of total domestic production (%)	Average yield/ha in tonnes	Potential yield/ha in tonnes
Rainfed upland	Ogun, Ondo, Abua, Osun, Ekiti, Oyo, Edo, Delta, Niger, Kwara, Kogi, Sokoto, Kebbi, Kaduna, FCT, and Benue	30	17	1.7	3.5
Rainfed lowland (FADAMA)	Adamawa, Ebonyi, Ondo, Ekiti, Edo, Delta, Rivers, Bayelsa, Cross River, Akwa Ibom, Lagos, all major river valleys	47	53	2.2	5
Irrigated	Adamawa, Niger, Sokoto, Kebbi, Borno, Benue, Kogi, Anambra, Enugu, Ebonyi, Cross River, Kano, Lagos, Kwata, Akwa, Ibom, Ogun	17	27	3.5	6-7
Deep water floating	Flooded areas: Rima Valley in Kebbu State and deep floofef areas of Delta State	5	3	1.3	2.5
Mangrove swamp	Ondo, Delta, Edo, Rivers, Bayelsa, Cross River, Akwa Ibom	1	1	2	4

Source: Ezedinma (2008).

stands at 87 km^3. Extractable ground water is 59.51 km^3 (FAO et al., 2014). The permeable (sedimentary acquifers) ground water is distributed in about 10 provinces in Nigeria, these provinces include: coastal alluvium, river valley alluvium, dahomy basin, kerri-kerri formation, Chad basin, sokoto basin, middle Niger basin, Anambara basin, Cross river basin and Benue basin (Matins, 2001). The Chad and the Sokoto basins lie along the northern international boundaries with sokoto representing Nigeria's segment of the internationally shared Iullemeden Aquifer System (IAS). To the south, Nigeria also has its share of ground waters- the trans-boundary costal aquifers of the Gulf of Guinea Tano and Keta Aquifer Systems (Goldface-Irokalibe, 2008). Water resources from external sources (republic of Niger, republic of Benin and the republic of Cameroon) make up an estimated 65.2 km^3/year of the surface water.

Inland, the country has very rich and abundant water resources with well drained land by rivers and streams some of which are seasonal (Gold-face, 2008; Takeshima and Adesugba, 2014; FAO, 2015). The surface water resource can be assessed by the major drainage basins covering various areas. These basins include; Niger basin, Lake Chad basin, the West Coast and West Central basins.

The country also has local ground water in shallow alluvial (FADAMA) acquifers adjacent to major rivers (FAO et al., 2014) and extensive ground waters in 8 hydrological areas namely: Niger North, Niger central, upper Benue, lower Benue, Niger south, western littoral, eastern littoral and the Lake Chad (FAO et al., 2014; Oyebande, 2015a). These are drained mainly by the River Niger, River Benue, Lake Chad and also the Oguta Lake and their several minor tributaries and rivers that discharge into them. Other perennial rivers that serve drainage include the Gongola, Hadejia-Jama'are, Kaduna, Zamfara and Yobe in the north, and the Ogun, Osun, Imo, Cross and Anambara rivers in the south (Goldface-Irokalibe, 2008).

The Niger River has an estimated annual drainage of 127 km^3 accounting for over 50% of the runoffs. The Niger River is shared by eleven countries and Nigeria benefits the most as 26% of its drainage lies within its boundaries (Matins, 2001). The river also houses the three major dams-

Table 2. Equipped and actual irrigation areas in Nigeria 2014.

Scheme type	Equipped area (ha)	Actual irrigated area (ha)	Actual irrigated as percentage (%) of equipped area
River basin development authorities	92,317	29,140	32
State schemes	12,200	6,700	55
Private sector- sugar schemes	5,600	0	0
Private small scale schemes	128,000	128,000	100
Improved FADAMA(equipped low land	55,000	55,000	100
Total	293,117	218,840	75

Source: World Bank (2014b).

the Kainji, Shiroro and Jebba (Martins and Olofin, 1992). The damming capacity of the country's water resources is estimated to be 45.6 km^3 (FAO et al., 2014).

There are a total of 200 dams in Nigeria as reported by Gold-face (2008) with 18 others (some of which are ongoing projects while others are in the pipe line to be constructed). Also, a total of other 83 water projects (ongoing and proposed) as reported by FAO et al. (2014) under the "Water for agriculture and energy: Nigeria". These projects are to expand irrigation to achieve the goals of the vision 20: 2020 aimed on transforming Agriculture to a sustainable profitable sector. These water projects include provision of boreholes, rehabilitation of existing dams and water reservoirs construction etc. These existing dams and ongoing irrigation projects are clear indications of irrigation development and are thus prospects for irrigation rice farming and self-sufficiency in rice production in Nigeria.

Also under the Agricultural transformation Agenda (ATA), (which also builds on the vision 20:2020) the federal government with the assistance of the world bank has proposed a rehabilitation of three major irrigation sites in the north; the Bakolori and Rima all in Sokoto states, Hadejia jama" are in Kano and the Dadin kowa in the old Gongola State. With so many large water bodies of lakes and water reservoirs Nigeria has the potentials to engage in irrigation farming.

However, the water resources of the country are reportedly being underutilized, for example in 2000 it was reported that the water withdrawal for agriculture is only 5.5 km^3. This could be explained by the findings of Valipour (2015a) that irrigation water requirement is affected by river basin management, water allocation policies and agricultural expansion which has earlier been discussed to be poor in Nigeria. Therefore, Nigeria needs the right policies for sustainable growth in the irrigation sector.

Land resource

The exact amount of land under irrigation in Nigeria is difficult to estimate because as observed by NINCID (2015), there is no commonly agreed definition of irrigation adopted by researchers and agencies. However, irrigation land potentials according to the estimates reported by FAO et al. (2014) vary from between 1.5 to 3.14 million hectares. According to this report, the potential irrigable land area is about 2.1 million ha. Of this, about 1.6 million ha is irrigable from surface water while 0.5million ha from ground water. Though, areas suitable for irrigation from ground water are yet to be assessed. However the World Bank (2014a) reported that areas which were equipped for irrigation at the FADAMA and for private small scale irrigation schemes are fully utilized while those for the RBDAs and sugar estate as well as the state development schemes are left underutilized. This situation shows potentials in land resources yet to be tapped.

About 150 new projects covering one million hectares of irrigable land, including dams, diver dams, inter basin water transfer and river training has been proposed for future implementation (Oyebande, 2015b). There are therefore, immense opportunities for prosperity for the Nigerian rice farmers and for development in the rice subsector (Table 2).

Higher returns over other ecologies

Irrigation is a technological input and productivity enhancing investment that stands out strongly as a result of its role in stabilizing yields (Sakairi, 2004). In addition to yield stabilization it avails the farmer the opportunity of multiple cropping in a year. Other advantages of irrigation farming that places it above other production systems is the fact that it increases food production and decreases

Table 3. Cost and returns structure to irrigation farming in Oshin irrigation scheme Nigeria (naira/ha).

Item	Averages
(a) Gross revenue (G.R) Less	76,833.61
(b) Total variation cost (TVC)	35,555.33
Seeds	3003.1
Agro-chemicals eg fertilizer, insecticide	10000.0
Hired labour	14773.2
Marketing/Transport Equals	7797.03
(c) Gross Margin (GM) Less	41278.3
Imputed interest on capital Less	1923.1
Imputed rental value of land Less	1500.0
Depreciations on hoes, cutlass, and other farm tools Less	2280.4
Imputed cost of family labour Equals	2034.9
Returns to Farmer's Labour and Management (RLM)	33539.5

Source: Fakayode et al. (2010).

the economic efforts of drought (Civil Engineers, 2015). This characteristics is of paramount importance in Nigeria in the face of climate change which has brought uncertainty to weather conditions in the country most especially in the northern part of the country which account for major food crops production in Nigeria e.g. cereals. Evidence from empirical studies has shown that higher yields are observed in irrigated crops particularly in rice. Average yields of 3.5 tonnes/ha has been observed in irrigated rice in Nigeria as against 2 tonnes/ha obtained in rain fed rice production even though these figures are far below observed yields of 6 tonnes/ha in other countries (Kebbeh et al., 2003).

These increases in yields are associated with a shift from traditional rice verities to modern varieties and also a shift in planting technique-from broadcasting to transplanting as well as increased in the use of other inputs like fertilizers (Mongkolsmai and Mark, 1989).

Irrigation has also been referred to as a driving force of agricultural income for farmers and the nation at large in that there is an opportunity for multiple harvests annually. Fakayode et al. (2010) in a study carried out at Oshin irrigation scheme in Kwara state reported that irrigation farming is profitable (Table 3). A farmer makes a return of about ₦33, 000 per hectare of irrigated rice. It is expected that since irrigation offers opportunity for multiple cropping a farmer will earn more income annually. These attributes of irrigation places it above other production ecology and offers high potentials for rice farmers and Nigeria as a whole towards the attainment of rice self-sufficiency and consequently food security in the near future.

CHALLENGES OF IRRIGATION IN NIGERIA

Policies and institutions

Policy fragmentation, implementation and inconsistency have been a major challenge in Nigeria and irrigation policies are not an exception. Water and agriculture policies have usually been developed independently by various government (formulating) agencies. Although the Federal Ministry of Water Resources (FMWR) has overall responsibility for formulating policies for irrigation development in Nigeria, the State Irrigation departments (SID) and River Basin Development Authorities (RBDAs) and the Federal Ministry of Agriculture and Rural Development (FMARD) also carry out their independent activities sometimes different from those of the FMWR. This situation has led to a fragmented and conflicting approach to irrigation development and resulted into a sort of competing rather than cooperating and complimenting approach (Goldface-Irokalibe, 2008; World Bank, 2014).

The World Bank (2014b) asserted that since the 1970s when the Federal government of Nigeria embarked on dam construction and large scale surface irrigation schemes, a number of policy documents have been produced. Only between 1998 and 2007, there were several policies and documents on irrigation which include; National Water Resources (NWR) policy, Draft of National Irrigation Policy, Water resources infrastructure operation and maintenance policy and financial report of the water resource strategy. Also, there is the NWR bill being currently prepared. This bill is in line with the global principles of Integrated Water Resources builds on much of the earlier works on irrigation and is aimed at addressing some of the irregularities of existing water policies in the country. The main component of the bill includes:

i. Assigning regulatory responsible to the Nigerian Integrated Water Resources Management commission (NIWRMC) an independent regulatory body and the water resource development to RBDAs.
ii. Promoting stakeholders participation in water

management such as Water Users Association (WUA).

However, except for a few countries like Egypt, Djibouti, and Ethiopia, Africa generally has an unfavorable/weak water management and land use policies (Valipour, 2014a). This weak framework has been a major challenge over the years particularly in Nigeria. A typical example is the RBDAs which according to Section 4(1) a-d of the RBDA Act has vested powers to construct and maintain irrigation dams and drainage system, supply water to all users, construct and maintain infrastructural services including roads and bridges in various project sites. Researchers (Goldface-Irokalibe, 2008; Worldbank, 2014) have identified inherent flaws in the Act and establishment of the institutions. Some of these flaws include RBDAs being simultaneous suppliers, regulators and consumers of water- engendering a conflict of interest. It has also been observed that rather than on hydrological boundaries, the operational domains of the country's RBDAs were chosen based on political boundaries. Goldface-Irokalibe (2008) argued that this political sentiments in delineating operational locations has led to RBDAs pursuing independent and single minded activities based on prevailing situations in their different local domains, thereby, resulting to lack of coordination between RBDAs and an unstable approach to water management. Another notable flaw is the empowerment of the RBDAs to supply water from their completed storage schemes to users for a fee to be determined by the concerned RBDA leaving the Authorities with no powers of recovering charges in any events of users defaulting.

The many policy documents and institutions all have common challenges which have led to somersaults in policies. Some of these challenges include the following:

1. A number of institutions have overlapping functions and duplication of mandates which are controlled by government with poor coordination. The result of this confliction is massive infrastructure procurement with little attention to operations and maintenance needs. And a lack of inter-sectoral coordination with each segment pursuing its independent agenda. This is so because of the institutional arrangement in the country's water resources (Goldface-Irokalibe, 2008) which has the Federal Government through the FMAWR (including 12 River Basin Development Authorities (RBDAs) and National Water Resources Institute (NWRI) – responsible for formulating and coordinating national water policies, development and management of large water resources infrastructure, dams, reservoirs, irrigation and water supply schemes.

The State Government on the other hand is vested with the responsibility of providing potable water through state Water Agencies (SWAs) while the Local Government level is responsible for provision of rural water supplies and sanitation facilities.

2. A weak stakeholder ownership and limited local autonomy. There has not been active participation of farmers who are the main users of irrigation water in policy formulation. Hence, there is not so much active involvement of users at the local level. Policy approach seems to be top-bottom with little or no community drive. Association such as the WUA has been weak from its inception and so could not carry out it functions of operating and maintenance of irrigation project. Although, participatory irrigation management is known to be effective it is yet to be institutionalized into the subsector. Therefore, participation of stakeholders continues to be weak. Invariably policies do not get to fulfill their intended purpose. WUA do not get sufficient incentives like better water delivery to keep paying and quite often farmers expect government to provide water for free.

3. Weak financial sustainability is another common challenge. This challenge has always resulted in some sort of short term investment on irrigation thus, lack of sustainability of irrigation schemes. Most often the Federal Government alone fully covers irrigation investment cost with very little or no contribution from State and Local Governments. Making the situation worse in that the recurrent cost of operation and maintenance are seldom recovered from farmers due to their refusal to pay for services which (in their opinion) are epileptic in nature (NINCID, 2015). This situation results in the inability of government to continue funding and stagnation in further cultivating the fields on the part of the farmers. This weak financial sustainability may be linked with short sightedness in maintenance and operation policies which has terribly reduced the economic life of irrigation facilities and consequently impaired water delivery capacities.

There is therefore a need for policy reforms and improvement on water service delivery to ensure a revitalized water subsector which will provide water to users and users in turn pay for the service rendered which will maintain a steady flow of revenue for continuous water supply and also reduce government financial burden.

Climatic change

Climate change is associated directly or indirectly with human activities which leads to global warming resulting in extreme weather conditions of drought, floods, storms etc. These extreme weather conditions as well as salt stress are the major problems associated with rice production (Ajetomobi et al., 2011) and are expected to worsen as time goes by subsequently affecting crop yields, productivity and food self sufficiency particularly in rice production (Odozi, 2014).

The rising temperature may increase or decrease water needs for agricultural use. The consequences of Sevier drought or flooding are likely to cause evapotranspiration resulting to water shortages in the dams- in cases of draught; while flooding will cause overflow of the dams resulting in the washing away of farmlands and crops

down streams, being that most of the irrigated lands particularly the small scale irrigation sites (FADAMA) in Nigeria are low lying- located on the flood plain (Bamidele et al., 2010). An overflow of the dams could result in colossal lost of farm land and crops particularly rice.

Climate change as suggested by Nzeadibe et al. (2011) requires the development of natural resource management strategies that ensures the suitable use of soil and water etc. Being that the large irrigation sites in Nigeria are poorly managed and maintained due to lack of a proper management system makes irrigation farming unattractive because they are prone to disaster at all times. Most of the canals and drainages are poorly or not managed. Valipour (2014c) in his study of pressure on renewable water resources in 2060 observed a variability in National Rainfall Index (NRI) and this he says can be as a result of climatic and evironmental factors like: green house gases, global warming and climate change. A typical example was in the year 2012 when farmers lost all their farmland and crops to flood from Lagodo dam in cameroon due to heavy rains recorded that year. It was reported that Nigeria lost a total of 2.6 trillion naira to the floods.

Low access to capital

Accessing credit has been one of the major problems farmers face in Nigeria. Minimal or non-existent of credit facilities to farmers particularly small scale farmers who form the bulk of individuals involved in the agricultural sectors has left the production system underdeveloped-characterized with low input use and low productivity. Without credit, farming, particularly rice will not move from its present extensive low subsistence production to a more intensive production (IFDC, 2008).

Irrigation farming has been known to be a high capital intensive production system in Nigeria. Consequently farmers are not encouraged to participate hence a possible reason for the low development of this production system.The following reasons are known to account for the high capital intensive nature of the irrigation system:

High level and cost of labour

Labour is an important indicator for irrigation farming and water management. Several studies have shown that labour could constitute a major setback in irrigation farming across the world in the future. According to Valipour (2013, 2014a, b, c, d; 2015a, b) the observed decrease in rural population and the economic population that is involved in Agriculture signifies a decline in labour availability in the future.

Mechanization in irrigation farming is also low in Nigeria, available production equipment are not suitable for use in areas prepared for irrigation (FADAMA) due to the soil type and the weight of these machines. Therefore, farmers resort to the use of manual labour which is unavailable and expensive. To lean credence to this, in a study by IFPRI (2014) on irrigation potentials in Nigeria conducted at the Bakolori irrigation scheme in Sokoto state, irrigated rice was reported to be labour intensive. Labour alone accounted for the largest cost component; a total of 250 mandays while bird scaring alone accounted for over 70 mandays at a cost of $4 per manday. This has the potential to discourage irrigation farming and also prevent the expansion of the existing farms.

In addition to the normal production activities of clearing, ploughing, weeding etc, farmers employ labour for draining and distribution of water for the small irrigation schemes and construction of canals and water channels for the large irrigation scheme. In some instances where the terrain of the irrigated sight are not accessible, most often farmers make personal efforts to provide access roads to their farms for easy transport of harvested crops or in the alternative employ labour to transport them.

High operating and development cost of irrigation

the cost of development of irrigation is quite high, as reported by FAO the average cost of developing irrigation in Nigeria in 2003 was $US 15000/ha while the average cost of maintenance of irrigation scheme in the 1990 stood at an estimated $US50/ha, $US 290/ha and $US 800/ha for gravity, pump and sprinkle irrigation types respectively. Operating cost in irrigations is also high. In small irrigation schemes where pumping machines are used farmers are constantly faced with irregular fuel supply and faulty machines. The cost of fueling and maintenance of these machines are high particularly in times of fuel scarcity when farmers are forced to patronize the black market buying at exorbitant prices. All of these lead to poor financial returns to farmers.

Availability and high cost of inputs

Irrigation farming demands the use of more farming inputs of seeds, fertilizers, pesticides and other chemicals as reported by IFPRI (2002) and Frolking et al. (1999) that Chinese farmers simultaneously increased the use of fertilizers and irrigation and fertilizer is known to work more effectively in the presence of sufficient water and soil (Li et al., 2004). The high cost and low availability of these inputs contributes to the high cost of production in irrigation farming.

Rice marketing

Marketing has been known to be an important aspect of production, the absence of a readily available market to

Table 4. Traders margin as percentage of retail prices, imported and domestic rice value chain (milled).

Margins	Domestic (%)	Imported (%)
Retailer margin	16	9
Wholesale trader margin	12	5
Initial rice trader margin	6	15
Farm gate price(paddy trader margin)	19	n/a
Total traders' margin	54	29

Source: Adopted from FAO (2013).

of farm products discourages farmers from producing and intensifying production. It is evident that the market for locally produced rice in Nigeria is characterized with low prices. The prices of locally produced rice are not competitive with rice imported from other sources. The price gap especially the very low farm gate prices offered for the product acts as a disincentive to farmers (FAO, 2013). Also the market structure and the policies of government in the rice sector which are meant to stimulate and improve local production are not yielding desired results. These policies have rather shown incentives to traders (FAO, 2013) who most times determine the prices at which to buy these produce. These traders are reported to have the higher margins in the domestic rice value chain as shown on Table 4.

FUTURE OF IRRIGATION IN NIGERIA

To ensure sustainability and viability in irrigation farming, several studies have tried to determine the future by identifying important factors that affect the sector. Attempts are made by these studies to estimate the effect these factors (socio-economic, demographic, climatic or political) and unfavorable land use policies have on irrigation. These estimates are used to project future status of irrigation in Nigeria.

To meet the expected demand for irrigation in Nigeria, rapid population growth and urbanization play a major and significant role (NINCID, 2015; Takeshima and Adesugba, 2014). According to NINCID (2015) urban population in Nigeria is expected to grow at an average rate of 4.2%, the increased proportion of the urban area is expected to reach 63% of the national population in 2025. A major change induced by urbanization will be an increase in the demand for food like rice and wheat. Assuming rice consumption grows at the same rate as the population, the demand for rice is expected to reach 11 million tonnes by 2025. About 2 million hectares mostly of irrigated land is required to produce such quantity. This requires an expansion of the irrigated lands which is expected to grow by 0.6% annually from 1995 to 2020 (NINCID, 2014). While this expansion is desirable, it is expected to put pressure on renewable water resources in the future (Takeshima and Adesugba, 2014, NINCID, 2015; Valipour, 2014c). Nigeria's abundant water resource is thus a potential to be meet this requirement by 2025. But effort will have to be doubled to develop the water resources to make the expected expansion in irrigated rice production feasible (NINCID, 2015).

On the other hand, urbanization has also been projected to reduce rural population and the economic population that is involved in agriculture. Movement of people from rural to urban areas will imply low labour supply in agricultural activities. Though Takeshima and Adesugba (2014) has reported abundant availability of labour in Nigeria, Valupour (2013; 2014a, b, c; 2015a, b) warns that labour availability in irrigation farming in future would generally be affected with the decline in rural population and the economic population involved in Agriculture.

Climate change, agricultural research and development effort are other constraints that are expected to limit irrigation expansion (Takeshima and Adesugba, 2014). The area cropped to permanent crop for example is a factor which is dependent on climatic conditions, farmers' tendency and government polices (which influences government policies). All these play an important role in the decision to allocate the required quantity of water for irrigation.

Other factors such as Human Development Index (HDI) and contribution of agriculture to Nations' GDP have significant effect on irrigation generally in Africa. Valipour (2014a) in his study on land use policy and agricultural water management of the previous half of the century in Africa based on 2011 FAO data, reported that the value of permanent crops to cultivated area, HDI and GDP were 8%, 0.471 and 31% respectively for Nigeria. However, Egypt having better agricultural water management than Nigeria has HDI of 0.662 and 22% as value of crops to cultivated area. This shows a low values for these important factors in Nigeria. An insightful look at the author's findings indicate that intense development of irrigation will earn farmers additional income which will in turn improve farmers welfare and raise agriculture contribution to GDP. Consequently, government will be better encouraged to invest in agricultural research and development which is a critical factor that will determine whether water is used more intensely in Nigerian

agriculture (Takeshima and Adesugba, 2014).

Finally, owing to the unfavourable land use policy in Africa particularly Nigeria, the future of private ownership which is believed to be an effective way of irrigation water management is bleak because no farmer would be willing to invest on a land which he owns no title or ownership. There is a therefore, a need for good and concerted government polices to encourage farmers to use irrigation system and raise cropping intensity for irrigated areas (Valipour, 2014a).

CONCLUSION AND RECOMMENDATION

This paper demonstrates that Nigeria has high potentials in rice irrigation farming particularly in areas of land and water resources which are relatively in abundance. Though yet to be fully developed, can afford the country the opportunity to attain rice self sufficiency in the nearest future. In addition to these important factors of production, irrigation also has higher yields potentials and offers opportunities for multiple cropping since water is guaranteed. Though, cost intensive, this production system brings reasonable returns to rice producers which boost the income of the farmers and the household.

To promote available and sustained rice irrigation production system, these bottle necks to increased irrigation rice production must be addressed. Amending some of these policies such as greater farmer participation in irrigation development should be encouraged; functions of water regulatory institutions should be streamlined with each institution given specific and defined roles to enhance efficiency in water resource management. Policies which also promote direct link of producer and consumers are necessary, this will provide protection for farmers ad avail them the opportunity of better prices for their produce. It is also important that assistance is given to farmers in the areas of subsidized inputs supply and well developed credit market for farmers to be able to access loans to purchase farm inputs as well as purchase and maintenance of farm equipment like the water pump. Another area of intervention which is of utmost importance is the purchase and even distribution of appropriate farm implement suitable for the irrigation ecology which will reduce the high cost of labour and the drudgery involved in manual labor employed in rice irrigation farming.

Conflict of Interests

The authors have not declared any conflict of interests.

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Review

Trichoderma- a potential and effective bio fungicide and alternative source against notable phytopathogens: A review

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Trichoderma, **is a useful, filamentous fungi and they are cosmopolitan in nature which have attracted the attention because of their multi prong action against various plant pathogens. There are several biocontrol mechanism employed by *Trichoderma against* plant pathogens, these include release of hydrolytic enzymes that degrade cell wall of phytopathogenic fungi, competition for nutrients, parasitism, and antibiotics. *Trichoderma* spp. generally grows in its natural habitat on plant root surface and therefore it controls root diseases in particular.**

Key words: *Trichoderma*, anatagonism, phytopathogens.

INTRODUCTION

Modern agricultural practices are getting affected by various problems such as disease, pest, drought, decreased soil fertility due to use of hazardous chemical pesticides, bio-magnification and various environmental problem. The fungal disease is one of the major constraints for agriculture productivity in India losses is about 22 to 25% of its crops due to fungal diseases each year. This damage is estimated to be Rs.50,000 crores annually. Although, use of fungicides in crop protection has certainly contributed for minimizing yield losses. So, indiscriminate and unscientific use chemical pesticides for prevention of various fungal disease leads to cause of resurgence of new pest, fungicidal resistance, ultimately affecting the land, soil fertility and remains toxic when consumed by humans and animals. Their application needs care and must only be used when the threshold limits exceeds. As a result, there is a need for some eco-friendly biocontrol agents that help in resolving problems. Hence, there is a need to look forwarded to develop practices and eco-friendly bio-control in order to reduce the dependence on synthetic agrochemicals. Therefore, recent trends favor the use of alternative substances derived from natural plant extracts, biological control and several others components. The various types of biological control agents such as bacteria and fungi are involved in biocontrol activity. Biological control is an essential part of these strategies as a substitute of agrochemicals.

Biological control agents (BCAs) offer an alternative to the chemical control of fungal phyto-pathogens as they can kill or limit the growth of pathogens without harming the host. Various bacteria, fungi and protists are known to

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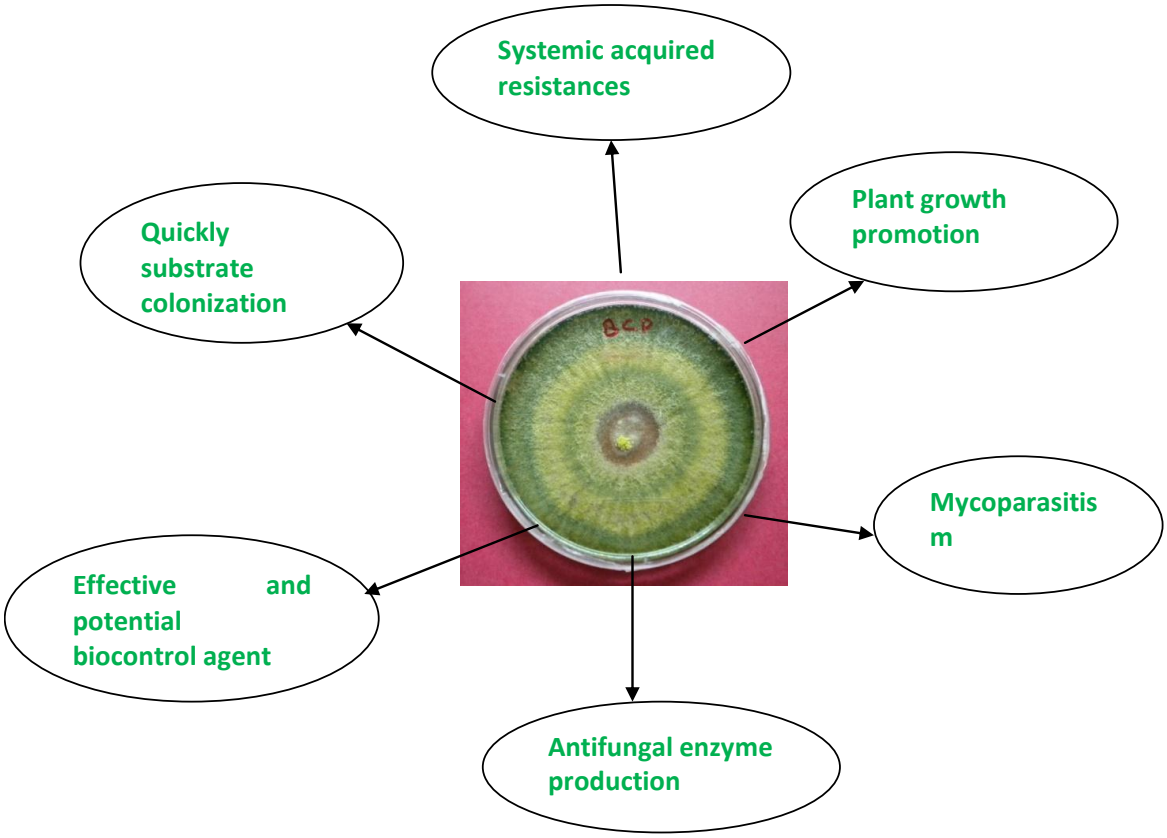

Figure 1. Biocontrol activities exhibited by *Trichoderma.*

have such features. They do so either by one or many of the following mechanisms: Competition for nutrients, secretion of lytic enzymes, secretion of toxic metabolites and direct parasitism on host (Agrios, 2005). Members of the genus *Trichoderma* have shown promise as BCAs (Elad et al. 1983; Chet 1987). The first practical control of crop disease with *Trichoderma* was in the control of root rot causing (*Armillaria mellea*) in citrus. It has since been very effectively used for the control of large number of soil borne plant pathogen like *Phytophthora, Rhizoctonia, Sclerotium, Phythium, Fusarium, Sclerotinia, and Galumannomyces.* Presently, entrepreneurs are mass producing its three main species viz, *Trichoderma harzianum*, *Trichoderma viride* and *Trichoderma koningii.*

They also have important economic impact, due to their ability to produce hydrolytic enzymes, secondary metabolites and degradation of xenobiotics (Harman et al., 2004). This versatile fungus is also used commercially in food and textile industry and as a source of transgene to develop disease resistance transgenic plants. This ecological and economic relevance of the genus *Trichoderma* makes it an ideal candidate as a model organism in various studies, spanning from molecular plant pathology, plant protection, biological control, and microbial ecology. Due to their rapid growth and capability of utilizing different substrates, species of this genus are often predominant components of the soil mycoflora in various ecosystems, such as agricultural, pasture, forest, salt marsh, prairie and desert soils over a wide range of climatic zones.

Trichodrma spp, native isolates must therefore be isolated and characterized for BCA activity so that they could be integrated into integrated pest management.

Fungal species belonging to the genus *Trichoderma* are worldwide spreads and easily isolated from soil by serial dilution technique. They are known as imperfect fungi, because they have no sexual stage. Faster growth rate in culture and production of conidia that are varying shades of green characterize fungi in the genus (Chaverri and Samuel, 2003). *Trichoderma* species are fungi with telomorphs belonging to the *Hypocreales* order of Ascomycotina division.

Trichoderma species are being shown to be the anamorphs of hypocrea species. Hypocrea was first described by Elias Fries in 1825 based on sphaeria rufa pers.: fr., aspecies with hyaline, ascospores.

In order to make the most effective use of biological control agent we must understand the mechanism of biological control. The various biocontrol mechanisms employed by *Trichoderma* are as follows (Figure 1):

1. Induced systemic resistances: Induced systemic

Table 1. Secondary metabolites release by *Trichoderma* species.

Name of Secondary metabolites	Release by	References
(Mannitol)	T. *hamatum*	hussain et al. (9175)
2- hydroxymalonic acid	T .*psuedokonigii*	kamal et al. (1971)
Trichodermaol	*Trichoderma species*	Adachi et al. (1983)
p-hydroxybenzyl alchol)	T.*koningii*	hung et al.(1995a)
1-hydroxy-3-methylanthraquinone	*T.viride PrL 2233*	slater et al. (1967)
Pencolide	T.*album*	Ren 1977
Viridiofungin A	*T.viride*ATCC74084	Harris et al. (1993)
Trichodermene A	*T .psuedokonigii*	Kamal et al. (1971)
Carbolic acid	*Trichoderma* species	Turner and Aldridge (1983)
Dermadin methyl ester	*T.polysporum*	Jin and jin (1989)
Isonitrin A	*T. harzianum* IMI 3198	Baldwin et al. (1991)
Spirolactone	*T.hamatum*HLX 1379	Baldwin et al. (1985a)
Dermadin	*T. viride* uc 4875	Pyke and Dietz (1966)

resistances (ISR) of plants against plant pathogens is a widespread phenomenon with respect to the underlying signaling pathways. After primary pathogen infection, plants are capable of producing an immune response which is known as systemic acquired resistances, the expression of pathogenesis gens such as acidic and basic β -1, 3 glucanase and chitinase and endochitinase are involved for the activation of SAR. The peptides, proteins and low molecular weight compounds produce by Trichoderma and these compounds induce ethylene production, hypersensitive response and other defence related reactions in plants.

2. Fungistasis: Most soils inhibit fungal germination and growth to a certain extent, a phenomenon known as soil fungistasis .*Trichoderma* grow rapidly when applied in soil, because they are resistant to many toxic compounds, including fungicides ,herbicides and pesticide such as DDT.

3. Production of cell wall degrading enzymes (CWDES): Chitinase, β -1, 3 glucanase produced by some *Trichoderma* species they are key enzymes in lysis of cell wall during their mycoparasitic action against plant pathogenic fungi.

4. Competition with other fungi: *Trichoderma* and other plant fungi feed on the waste products of the plant. Because *Trichoderma* can colonize the roots very rapidly it has a benefit in the competition for these nutrients.

5. Mycoparasitism of other fungi: *Trichoderma* can parasitize other fungi and release a digestive enzyme which breaks down the chitin in these fungi. In this way nutrients will be extracted from theme.

6. Secondary metabolites: They are producers of extracellular proteins, and are best known for their ability to produce enzymes that degrade cellulose and chitin. They also produce other useful enzymes such as endochitinase, chitinase , glucanase cellulase play a an importent role in biocontrol activity. Different strains produce more than 100 different metabolites that have known antibiotic activities. It is found that large variety of volatile secondary metabolites produced by Trichoderma such as ethylene, hydrogen cyanide, aldehydes and ketons which play an important role in controlling the plant pathogens (Table 1).

MAAS PRODUCTION OF *TRICHODERMA*

In India talc based formulation was first of all developed in Tamil Nadu Agriculture university, Coimbatore (Jeyarajan et al., 1994). Talc based formulation is formed by both solid and liquid state fermentation.

Solid state fermentation

This is the most common method for *Trichoderma* mass production. Several grains like sorghum, corn, rye and millets are used as substrates for *Trichoderma* mass production. These grains are moistened, sterilized and inoculated with *Trichoderma* spp and incubated for 10 to 15 days. *Trichoderma* produces dark green coloured spores covering the whole grain. These grains can be powdered and mixed in sterilized talc powder in 1:9. The finally processed product can be used for soil and seed treatment. Solid state fermentation is used for commercial level production; the main disadvantage associated with this technique is that it is very tedious and time consuming.

Liquid state fermentation

In liquid fermentation system *Trichoderma* is grown in liquid medium in shaker and ferment or and after the incubation period completes the fermented media is mixed in sterilized talcum poder in 1:9 ratio. Maximum

Table 2. Growth media used for production of *Trichoderma* in liquid state fermentation.

Growth media	References
Molasses and brewers yeast	Sankar and Jeyarajan (1996)
Potato dextrose broth, V -8 juice and Molasses-yeast medium	Prasad and Rangeshwaran (1998)
Molasses soy medium	Prasad and Rangeshwaran (2000)
Jaggery-soy medium	Prasad et al. (2002)

Table 3. Agro domestic wastes used to produce *Trichoderma* sp. by solid state fermentation.

Substrate	References
Sorghum grain	Upadhyay and Mukhopadhyay (1986)
Wheat bran	Mukhopadhyay (1986)
FYM,Rice straw	Kousalya (1990), Sangeetha et al. (1993)
Spent compost of mushroom	Singh et al. (2012)
spent tea leaf waste and coffe husk	Bhai et al. (1994)
Decomposed coconut coir pith spent malt	Gopalkrishanan et al. (2003)
Neem cake + wheat flour 10%	Singh et al. (2012)

biomass of *Trichoderma* can be produced in short time by using appropriate fermentation media and fermentation conditions such as aeration, agiatation, temperature, ph and antifoam control. Project Directorate of Bilogical Control (PDBC) Banglore revealed that maximum amount of biomass and viable propagules of *T. harzinum/ T. viride* can be obtained within 96 h of fermentation in a fermentor with aeration, agitation, temperature control (Prasad and Rangeswran 1998) (Table 2)

AGRO DOMESTIC WASTES AS SUBSTRATES FOR THE MASS MULTIPLICATION OF *TRICHODERMA*

For mass production the first step is to identify a suitable substrate which should be cheap, stable and easily available. The various agro domestic wastes used for *Trichoderma* mass production are Corn flour, sorghum grain, wheat bran, farm yard manure, Tea waste, wheat straw rice bran vegetable waste.

Various agricultural and domestic wastes such as: Corn flour, sorghum grain, wheat bran, farm yard manure, tea waste, wheat straw, rice bran, vegetable waste, had been evaluated for the mass multilplication of Trichoderma (Table 3). For formulation purpose all the waste materials are dried and grounded to a fine powder and after this they are filled in autoclavable bags. The materials were subjected to 45% of moisture and sterilized twice at 121°C. Seven day old culture of *Trichoderma* sp. was used to inoculate the waste medium. The spore suspension of the *Trichoderma* in DDW was made (1×10^6 conidia per ml). The plastic bags were then inoculated with the 10 ml of *Trichoderma* spore suspension and incubated at 28°C. After four days of incubation the inoculated bags were transferred in clean and sterilized plastic trays and the trays were covered with thin plastic film. After ten days of incubation the substrate is mixed with sterilized talcum powder and CFU of *Trichoderma* was calculated on every substrate (Figure 2).

SELF LIFE OF *TRICHODERMA* FORMULATIONS

Shelf life is the most important factor which determines the quality of *Trichoderma* bioformulation. Talc, peat, lignite and kaolin based formulations of *Trichoderma* have shelf life of 3 to 4 months. CFU count of *Trichoderma* in talc formulation has been found to reduced to 50% after 120 days. At PDBC in Bangalore, it was found that the shelf life of *Trichoderma* formulation was increased using various ingredients (chitin and glycerol) and heat shock at the end of log phase of fermentation was carried out which extends the shelf life of talc based formulation up to one year (Sriram et al., 2010; Sriram et al., 2011).

EFFECT OF THE ADDITION OF COLLOIDAL CHITIN EXTRACTED FROM SEA SHELLS ON THE SHELF LIFE OF *TRICHODERMA SP.*

To study the effect of colloidal chitin in on the shelf-life of talc formulations of *Trichoderma* sp. Colloidal chitin was prepared and added at the rate of 1, 2 and 3% (w/v) in PDB (Potato Dextrose Broth) and Sorghum Based Medium used in liquid as well as solid state fermentation. And CFU was counted regularly to check the effect of colloidal chitin on shelf life of *Trichoderma* bioformulation.

Figure 2. Effect of different agro and domestic based substrates on the population of *Trichoderma harzianum* (Th. Azad).

Figure 3. Different Concentration (1, 2 and 3%) of colloidal chitin in potato dextrose broth base medium.

Addition of colloidal chitin at 2% in production medium enhanced the shelf-life as compared to control (Figures 3 and 4)

Application of *Trichoderma* formulations

Several methods are recommended for the application of Bio fungicides for the successful management of plant diseases. Delivery and establishment of *Trichoderma* (bio fungicide) also play a very important role to the site of action (Table 4). The most common methods of application of *Trichoderma* are, by seed bio priming, Seedling dip, soil application and wound dressing, and foliar spray is rare.

Seed treatment

Seed treatment is one of the easy and effective method. In seed treatment seed is coated with dry powder/ dusts of *Trichoderma* just before the sowing. For commercial purpose, dry powder of antagonist is used at 3 to 10 g per kg seed based on seed size (Mukhopadhyay et al., 1992). *T. harzianum, T. virens* and *T.viride* were found to be effective seed protectants against *Pythium spp*. and *Rhizoctonia solani* (Mukherjee and Mukhopadhyay, 1995).

Seed biopriming

In this method seeds are treated with *Trichoderma* formulation and incubated under warm and humid conditions just prior to the emergence of radical. This technique has potential advantages over simple coating of seeds as it results in rapid and uniform seedling emergence. *Trichoderma* conidia germinate on the seed surface and form a layer around bioprimed seeds. Such seeds tolerate adverse soil conditions better.

Root treatment

Seedling roots can be treated with spore or cell suspension of antagonists either by drenching the bioagent in nursery beds or by dipping roots in bioagent suspension before transplanting. This method is generally used for the vegetable crops, rice where transplanting is practiced (Singh and Zaidi, 2002).

Soil treatment

There are several reports on the application of biocontrol agents to the soil and other growing media either before or at the time of planting for control of a wide range of

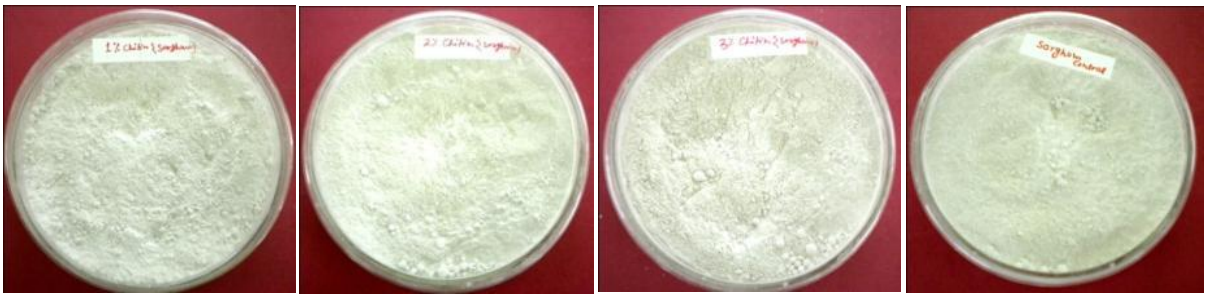

Figure 4. Different concentration (1, 2 and 3%) of colloidal chitin in sorghum base medium.

Table 4. *Trichoderma* species and its uses against different plant pathogens.

Bioagent	Plant pathogen	Crop
T. *viride*, T. *longibrachiatum, T. virens*	*Pythium*	Ginger
T. viride, T. *harzianum*	*Colletotrichum*	Chilli
Trichoderma koningii	*Ustilago segetum var tritici*	Wheat
T. *harzianum*	*Phytophthora*	Cardomom
Trichoderma spp.	*Cercospora moricola*	Mulberry
T. viride, *Trichoderma* species	*Fusarium udum*	Pigeon pea
T. *viride*, T. *harzianum*	*Botrytis cinerea*	Rose

soil-borne fungal pathogens (Baby and Manibhushanrao, 1996). Such applications are ideally suited for greenhouse and nursery**.**

Aerial spraying/wound dressing

Trichoderma has been successful applied to the aerial plant parts for the biocontrol of decay fungi in wounds on shrubs and trees (Papavizas, 1985).

CONCLUSIONS

Bio fungicide is one of the best alternatives against some notable plant pathogens. The limitations to biocontrol use are due to lack of knowledge on the ecology of the rhizosphere and use of in vitro antagonism for selection of biological control agents. But, the advantages of this method are more. *Trichoderma* spp. that are common saprophytic fungi found in soil, have been investigated. And it has been emerged as a as potential biocontrol agents because of their ability to reduces the incidences of disease caused by plant pathogenic fungi.

Conflict of Interests

The authors have not declared any conflict of interests.

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Soil moisture maintenance methods in cultivation in a greenhouse

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Soil water content standardization in agricultural experimentation is essential to mitigate variability within treatments, decreasing experimental errors arising from irrigation management. The aim of this study was to assess soil moisture maintenance methods using sunflower (*Helianthus annuus* L.) grown under controlled conditions as indicator plant. The experiment was conducted in a greenhouse during the period between August and October 2014, using an oxisol. Treatments consisted of four soil moisture maintenance methods (tensiometer, Irrigas®, gravimetric method and self-watering system) in eight repetitions, using an entirely randomized design. Variables analyzed were: chlorophyll content, dry matter of the head, leaves, stem, root, and total dry matter, water consumption and water use efficiency for head dry matter yield. Irrigation management based on gravimetric method is sufficient to provide higher biomass accumulation in the crop, consequently increasing plants water consumption.

Key words: Soil water content, *Helianthus annuus* L., gravimetric method.

INTRODUCTION

In tests under controlled conditions, soil water availability is a limiting factor to the growth and development of plants. Thus, the use of soil moisture maintenance methods is necessary to reduce variability within treatments, ensuring statistical tests sensitivity, and consequently, higher reliability to the results (Beltrão et al., 2002).

However, in general, soil moisture maintenance in agricultural experiments, particularly in those conducted in a greenhouse, is done without the adoption of criteria to standardize the water flow restored to experimental units.

Soil moisture variation study requires the use of appropriate methods when monitoring plants water availability (Silva et al., 2012), to prevent excess or drought conditions.

One of the most widely used methods for soil moisture maintenance in scientific experiments is conducted in vases using the gravimetric method. Using vase capacity characterization, also known as pot capacity, soil water volume is kept through the weighing difference of experimental units in a given time interval (Bonfim-Silva et al., 2011).

Taking into consideration the probable soil water

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content variations, other methodologies were proposed for irrigation control in experiments, among which self-watering system (Silva et al., 2005), Irrigas sensor (Calbo and Silva, 2005; Silva et al., 2014) and tensiometer (Richards, 1942; Libardi, 2005) are highlighted, the last two being identical to the commercially available.

Studies have proven that sunflower is one of the most sensitive crops to soil moisture variation, being used as indicator for plant of water availability level. Nazarli et al. (2010) and Dutra et al. (2012) demonstrated the close relation between soil moisture and crop yield.

Taking into account the different plant experimental treatments conducted in pots, moisture standardization is essential for all experimental plots, to obtain answers between applied treatments and to minimize the coefficient of variation errors during the experimental period.

In this context, it is necessary to establish soil water content maintenance methods that do not affect the yield of plants under experimentation, ensuring higher results reliability. Therefore, the aim of this study was to assess soil moisture maintenance methods using sunflower (*Helianthus annuus* L.) as indicator plant, under controlled crop conditions.

MATERIALS AND METHODS

The experiment was carried out in a greenhouse at the Agricultural Engineering Graduate Program, Federal University of Mato Grosso, municipality of Rondonópolis, Mato Grosso state. The city of Rondonópolis is located at 16° 27' 52" South latitude and 54° 34' 46" West longitude, at an altitude of 290 m above sea level.

The experimental design was completely randomized, with four soil moisture maintenance methods (gravimetric method, Irrigas sensor, tensiometer, and self-watering system) and eight repetitions, in the period between August and October, 2014.

An oxisol soil collected at 0 to 0.20 m depth in a Cerrado area was used. The soil was passed through a sieve with 4 mm mesh.

Soil characterization was conducted in accordance with EMBRAPA (1997), having the following physical and chemical characteristics: pH ($CaCl_2$) = 4.1; P (Mehlich) = 2.4 mg dm^{-3}; K = 28 mg dm^{-3}; Ca = 0.3 $cmol_c$ dm^{-3}; Mg = 0.2 $cmol_c$ dm^{-3}; H = 4.2 $cmol_c$ dm^{-3}; Al = 1.1 $cmol_c$ dm^{-3}; SB = 0.6 $cmol_c$ dm^{-3}; CTC = 5.9 $cmol_c$ dm^{-3}; V = 9.8%; Organic Matter = 22.7 g dm^{-3}; Sand = 549 g kg^{-1}; Silt = 84 g kg^{-1}; Clay = 367 g kg^{-1}.

Experimental units were represented by plastic pots with 3.18 dm^{-3} capacity. Base saturation was raised to 60% with dolomitic limestone (PRNT = 80.3%) addition, being kept for 30 days in order to decrease soil acidity.

After limestone incubation, fertilization with phosphorus and potassium was conducted at doses of 150 and 100 mg dm^{-3}, respectively, using single superphosphate and potassium chloride as sources.

Nitrogen fertilization (200 mg dm^{-3}) was split in three applications, at 10, 20, and 40 days after emergence (DAE). Micronutrient fertilization was conducted using a solution containing 1 mg dm^{-3} of B and Cu, 3 mg dm^{-3} of Mn and Zn and 0.2 mg dm^{-3} of Mo, provided by the following sources: boric acid, copper chloride, manganese chloride, zinc sulfate, and sodium molybdate, respectively

The cultivar used was Helio 250. Seeds were treated with systemic and contact fungicide. Sowing was conducted by placing ten seeds per pot, and 18 days after emergence (DAE), thinnings were carried out, leaving the two most vigorous plants.

The average daily maximum temperature observed during the experiment was of 36.7°C, and average values of maximum and minimum relative humidities were, respectively, 87.7 and 30.58%.

For irrigation management on soil water maintenance methods, van Genuchten (1980) equation was used according to the determination of the water retention curve in the soil (Dourado et al., 2000), with undisturbed soil samples being taken from experimental units (Equation 1).

$$\theta = \frac{0.468}{[1 + (0.0573|\Psi m|^{0.3545})]^{0.5724}} \quad (1)$$

where θ is the humidity (cm^3 cm^{-3}), and Ψm is the matric potential (cmH_2O).

Irrigation of all treatments was carried out daily at 7 a.m. and 2 p.m. For soil water content maintenance, an analog scale was used to replace water by the gravimetric method (Figure 1A), and a digital tensiometer was used to measure water tension in the soil (Figure 1B).

The gravimetric method for soil water retention maximum capacity maintenance, or pot capacity (PC), was determined in the laboratory in 3.18 dm^3 pots containing the equivalent of 3.620 kg soil at 3% moisture with mass basis, in ten repetitions. Thus, pots were placed on a support within plastic trays. Water was added up to two-thirds the height of the pots, so that the soil was saturated by capillarity, expelling all the air contained in its pores (Bonfim-Silva et al., 2011).

After soil saturation, pots were removed from the tray and placed on a support to allow free water draining. When pot drainage stopped, deformed soil samples were taken for mass basis moisture determination.

After samples removal, they were immediately weighed to obtain the wet weight. Afterward, they were led to a forced air oven at 105°C for a period of 24 h. After this period, samples were weighed again, and moisture regarding pot capacity was obtained by difference.

With a view to irrigation management during the crop cycle, the volume of water needed to maintain soil moisture at 80% PC was calculated. Thus, water volume and dry soil matter were added to the pot weight, in order to obtain the total weighing value at the time of experimental plots irrigation.

Tensiometry irrigation management was conducted based on soil water tension assessment, using a digital tensiometer (Figure 1B). The water volume to be applied by irrigation was calculated based on the water retention curve in the soil. The ideal water tension in the soil for moisture maintenance was set as 5 kPa.

After tensions had been observed, the corresponding moistures were calculated through van Genuchten (1980) equation. Knowing the current soil moisture and volumetric moisture at 80% PC, water replacement volume was calculated (Equation 2). Water application was conducted manually by measuring the water volume in a semi-analytical balance.

$$V = (\theta_{fc} - \theta_{actual}) \times 3180 \quad (2)$$

Where V is the water volume (cm^3), θ_{fc} the humidity at 80% field capacity, θ_{actual} the humidity in each treatment tension (cm^3 cm^{-3}) and the value 3180 is the soil volume in the pot (cm^3).

Irrigation control Irrigas type signaling system was also used to manage soil moisture in the experimental units (Calbo and Silva, 2005). These sensors were manufactured with a wide critical tension range, making the bubbling pressure test necessary, as described in Libardi (2005), in order to standardize porous capsules, selecting those with the same critical tension. In addition, the test was also used to verify if there were bubbling and air

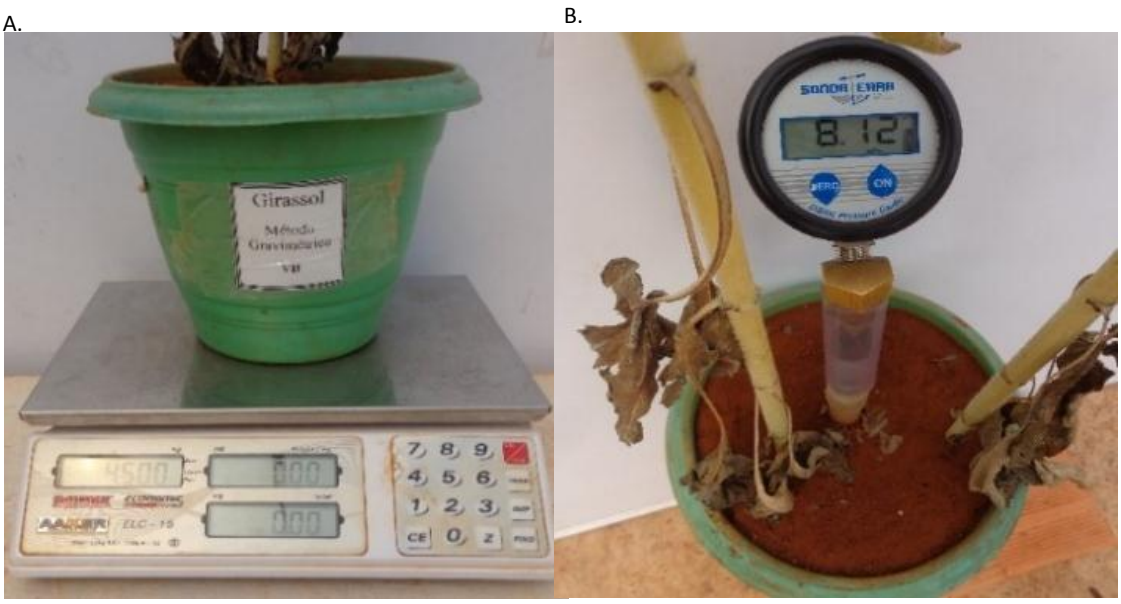

Figure 1. Irrigation management in (A) gravimetric and (B) tensiometer methods.

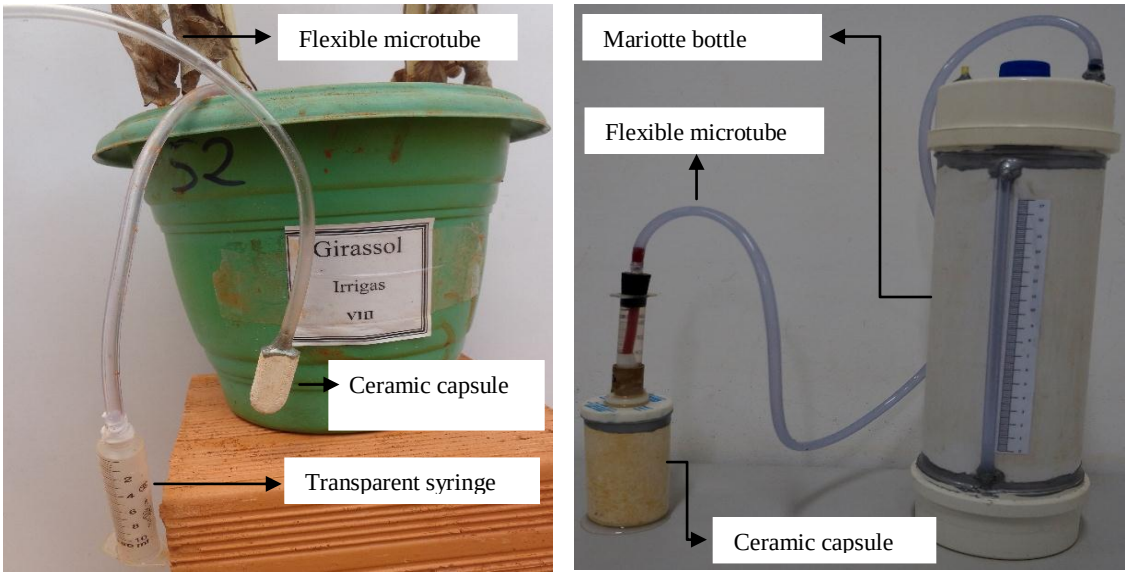

Figure 2. (A) Irrigas sensor and (B) self-watering system components.

leakage at Irrigas system seams.

Porous capsules were installed in the pot at 15 cm depth, in the wider scope of the root system region. Irrigation time was determined from 20 kPa tension, applying a volume of 250 ml water in each pot, calculated according to Van Genuchten equation. Irrigas system was composed of a sensor (porous capsule) connected through a flexible microtube to a transparent syringe (3 ml), which served as the system signaling device (Figure 2A).

Soil moisture in the self-watering system treatment was maintained with continuous water replacement, in accordance with plant needs. The system consisted of a porous ceramic capsule (filter candle with 0.05 m diameter and 0.07 m height) inserted into the soil through the upper portion of the pot. A flexible microtube (0.005 m inner diameter and 0.001 m wall thickness) connected the ceramic capsule to the constant level reservoir (Mariotte bottle), which was located below the pot (Figure 2B).

Water potential in the soil was established by the water column height between the pot and the reservoir (30 cm), corresponding to a controlled tension of 3 kPa. Thus, continuous crop evapotranspiration ensured water automatic replacement to the soil, making it a self-watering system (Silva et al., 2005). A level scale calibrated in mm and attached to the reservoir enabled water consumption measurement in each experimental unit.

Experiment collection was conducted 69 days after emergence,

Table 1. Chlorophyll Index of sunflower plants subjected to soil moisture maintenance methods*.

Stage	Vegetative		Reproductive		
Methods	15 DAE	22 DAE	50 DAE	57 DAE	69 DAE
Gravimetric	35.63^{b}	39.34^{a}	32.04^{b}	35.05^{ab}	32.53^{ab}
Tensiometer	36.49^{b}	39.44^{a}	33.35^{ab}	35.30^{ab}	34.20^{a}
Irrigas	38.94^{a}	39.43^{a}	34.41^{a}	35.74^{a}	36.10^{a}
Self-watering	36.24^{b}	36.34^{b}	31.81^{b}	32.59^{b}	28.68^{b}
CV (%)	4.54	3.43	3.65	6.33	9.88
Means	36.82	38.63	32.90	34.67	32.88

*Means followed by the same lowercase letter in the column do not differ by Tukey test at 0.05 probability.

and chlorophyll content, dry matter of leaves, stem, capitulum, roots, and total dry matter were assessed. In order to obtain the dry matter, samples were packed in paper bags and placed to dry in a forced air circulation oven for 72 h at 65°C, until obtaining a constant weight.

In order to obtain the chlorophyll index, portable chlorophyll index electronic meter (Falker ClorofiLOG® CFL 1030 model) was used. Through chlorophyll meter, reading per leaf was conducted in five middle third leaves located on the main stem of sunflower plants, obtaining a chlorophyll index average per experimental unit.

In addition to these variables, the volume of water consumed during the experimental period and water use efficiency in relation to productivity were also assessed. Results were submitted for analysis of variance when significant to the Tukey test at 5% probability by Sisvar Statistical Program (Ferreira, 2011).

RESULTS AND DISCUSSION

Significant differences were observed between the statistical methods analyzed for chlorophyll index when comparing the different assessment periods. The maximum value of 39.44 was obtained at 22 DAE, decreasing during the other assessments to the minimum value of 28.68 after 69 DAE (Table 1).

Chlorophyll index

Chlorophyll index decrease can be explained by the plant being in the reproductive stage at 34 DAE, period in which photo assimilates were transported to form inflorescences, causing leaf chlorophyll content to decrease.

Dordas and Sioulas (2008) found chlorophyll content decrease from the anthesis stage (65.50) to the stage of plant physiological maturity (49.94). The authors attributed this decrease to nitrogen relocation in the plants, with this nutrient being remobilized to form and develop achenes.

Observing the effect of treatments at 69 DAE (harvest), it was found that chlorophyll index in the Irrigas method exceeded by 20.55% the value obtained in the self-watering system, which provided soil moisture constant maintenance.

Higher chlorophyll indexes observed in the Irrigas system corroborated with Oraki et al. (2012), who reported that, in lower water availability conditions, chlorophyll content increase was observed in all sunflower hybrids analyzed, averaging 49.02 in the treatment maintained at the lowest water content in the soil.

On the other hand, Dutra et al. (2012) observed chlorophyll content decrease in sunflower plants of treatments with higher water availability in the soil, resembling the self-watering system. In this case, the higher water content in the soil caused leaf senescence increase by increasing ABA and ethylene concentration, inducing chlorosis increase due to chlorophyll degradation.

In general, assessed productive characteristics highest averages were observed in the gravimetric method, except for the water use efficiency variable, which had higher values in the Irrigas system.

Dry matter

For leaf dry matter, the self-watering system and the gravimetric method promoted the highest averages in relation to tensiometer and Irrigas methods (Table 2).

Sunflower plants showed a positive response to soil moisture maintenance by the self-watering system for this variable. Results may imply that self-watering system constant irrigation provided a condition in which stomata remained open for a longer period of time, establishing higher CO_2 amounts, which is responsible for increasing the dry matter of leaves in plants subjected to this treatment (Nazarli et al., 2010).

Canavar et al. (2014) reported the positive influence of soil water maintenance (30 and 60% field capacity) by the gravimetric method in leaf dry matter in an experiment with four sunflower genotypes under controlled conditions at 50 days after sowing.

Sunflower plants showed higher stem dry matter when subjected to the gravimetric method. It was also noted that other methods, such as tensiometer, self-watering system, and Irrigas have not promoted significant differences for this variable (Table 2). Possibly, higher

Table 2. Dry matter of leaves - DML, stem - SDM, Capitulum - DMCAP, root - DMR and total dry matter-TDM (g plant^{-1}) of sunflowers cultivated with soil moisture maintenance methods at 69 days after emergence (DAE)*.

Variable	Gravimetric	Tensiometer	Irrigas	Self-watering	CV (%)
DML	13.48^{a}	11.92^{b}	10.80^{b}	13.85^{a}	8.34
SDM	19.09^{a}	15.67^{b}	14.60^{b}	15.58^{b}	11.96
DMCAP	19.53^{a}	18.97^{a}	19.11^{a}	12.43^{b}	11.13
DMR	11.76^{a}	7.87^{b}	7.70^{b}	8.79^{b}	20.56
TDM	63.86^{a}	54.43^{b}	52.21^{b}	50.65^{b}	8.56

*Means followed by the same lowercase letter in the line do not differ by Tukey test at 0.05 probability

water consumption in the gravimetric method induced sunflower photosynthesis increase, followed by photosynthetic area growth (Hemmati and Soleymani, 2014) and stem biomass production increase.

Boareto et al. (2012), while researching evaporation levels variation effect for this crop, reported that lower water content in the soil promoted plant water stress, reducing evapotranspiration due to lower stomatal conductance. This process substantially lowers carbon assimilation and stem biomass production.

For head dry matter, the lowest average was found in the self-watering system (12.43 g), being 36.35% lower when compared with the value obtained in the gravimetric method (19.53 g). There was no statistical difference between the other methods (Table 2). Soil moisture maintenance by the self-watering system may have influenced in the nitrogen assimilation by plants, influencing biomass accumulation in head formation and development.

Zobiole et al. (2010) mentioned that, in head and achene formation, there is photosynthate translocation from accumulated reserves, mainly in sunflower plants leaves and stems, which are dependent on nitrogen availability to the plant. Among soil moisture maintenance methods, it was found that the highest root dry matter yield was observed in the gravimetric method (11.76 g pot^{-1}), which was statistically higher than the others (Table 2).

The result can be explained by root distribution in the pots, once plants subjected to the self-watering system focused higher root matter near the porous capsule. Gravimetric method, on the other hand, distributed the water volume evenly in the whole pot, so that the root system exploited higher soil volume, promoting higher root biomass.

Paiva et al. (2011) found a significant effect on sunflower plants root system development when they were exposed to water levels in the soil. In addition, the highest root dry matter value was obtained in the treatment equivalent to 75.17% field capacity, corroborating the results of this study, in which the gravimetric method promoted better root development. In sunflower plants total dry matter, the highest yields were observed in the gravimetric method (Table 2).

For Farahvash et al., (2011), the sunflower plants total dry matter decrease is the result of drought during the period of plant growth and development, causing leaf area and photosynthetic process reduction, besides reducing photoassimilates production and leaf, stem, and head development.

Nobre et al. (2010) observed 140% shoot dry matter increase when comparing the hypoxic condition with 80% crop water requirement replacement, corroborating the results of this study, in which the highest biomass yields were found in the gravimetric method, where soil moisture was maintained at 80% field capacity.

Water consumption and use efficiency

There was a significant variation in water use by sunflower plants depending on the methodology used to maintain soil moisture. Among methods, the highest water consumption accumulated in the period of 69 days was of 27.39 L in the gravimetric method, which statistically differed from the values verified in the other methods, except for the tensiometer (Figure 3).

It should be noted that, among treatments, only irrigation held in the gravimetric method was not based on water tension in the soil. Instead, it was based on bulk water content, which supported moisture content increase in pot capacity (PC).

Souza et al. (2000) attributed soil available water superiority to the gravimetric method based on PC determination, since the soil, after being saturated, only undergoes potential gravitational action. Gravitational potential action power is certainly less expressive than the 5 kPa tension (Porto et al., 2014), present to the other methods based on water tension in the soil, what provided soil water volume increase in PC, and higher water consumption.

Flénet and Saraiva (1996), while investigating sunflower crop response to soil water contents, also observed a significant effect of water consumption by plants, reporting higher consumption in the treatment that held the soil under field capacity.

Water use efficiency showed the significant effect to head dry matter when subjected to soil moisture

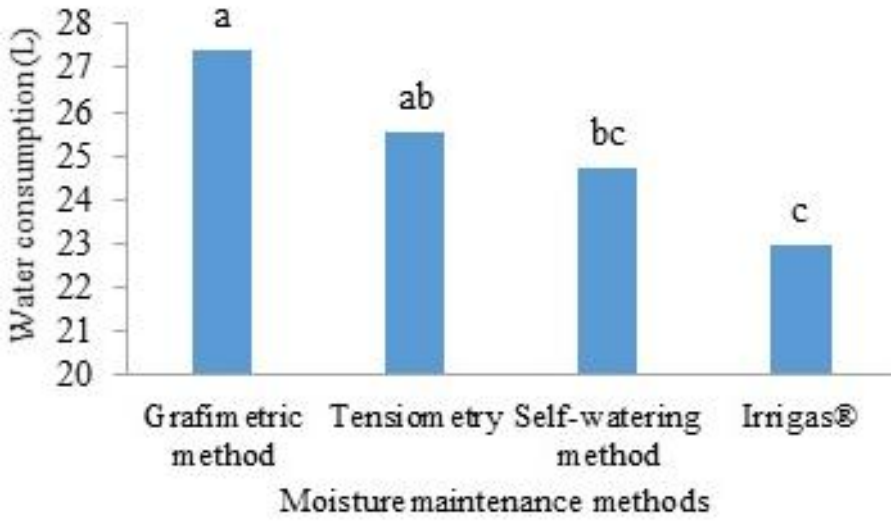

Figure 3. Water consumption of sunflower cultivated with soil moisture maintenance methods at up to 69 days after emergence.

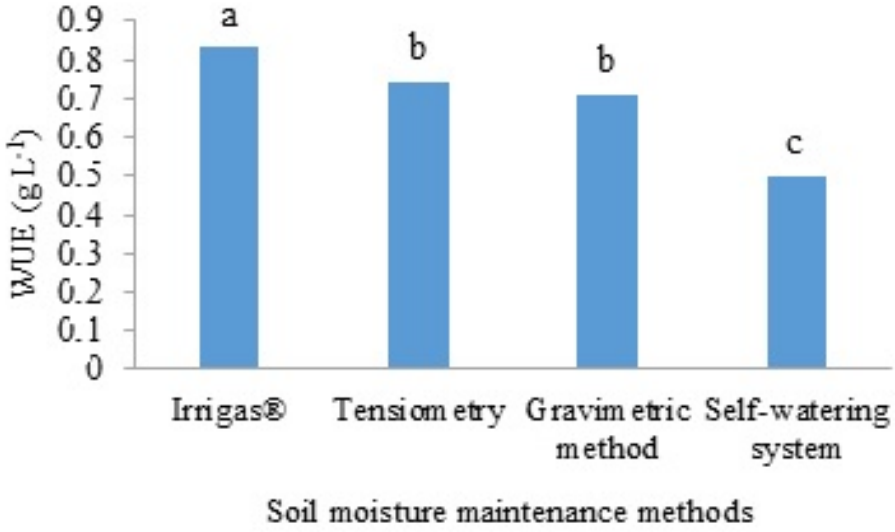

Figure 4. Water use efficiency for head dry matter (WUE) of sunflower cultivated with soil moisture maintenance methods at 69 days after emergence

maintenance methods. It was noted that Irrigas (0.83 g L^{-1}) had the highest average, with a 39.76% increase when compared with the self-watering system (0.50 g L^{-1}) (Figure 4).

This effect may be related to nutrients accumulation by plants subjected to irrigation management with Irrigas sensor, thus influencing head dry matter yield. Thus, higher nutrient concentrations provided further head development.

Results, combined with lower water consumption indicated by Irrigas sensor, caused this method to obtain the best water use efficiency, providing 0.83 g head dry matter production for each 1 L of consumed water.

Steduto and Albrizio (2005) have also observed a significant effect of water content variability in the soil on water use efficiency in sunflower plants. They defined water use efficiency as a parameter that relates carbon assimilation by plants to evapotranspiration accumulated at the end of the cycle, constituting a characteristic of great importance in crop biomass gain assessment.

In addition, Naim and Ahmed (2010) reported water use efficiency significant effect for sunflower achenes production in function of different water availabilities in the soil, in which the treatment with the lowest plant water

consumption had the highest water use efficiency.

Conclusions

The correct choice of soil moisture maintenance method in basic research, as tests in the greenhouse, is of great importance for the results obtained, and is a result of the treatments and never the inappropriate management of moisture, once the production of sunflower was influenced by the methods of soil moisture maintenance. When applying the gravimetric method is most required volume of water. The lowest values of production parameters were found when the maintenance of soil moisture was performed with Irrigas sensor. The management of sunflower irrigation based on the gravimetric method is sufficient to provide the greatest accumulation of biomass and therefore increased consumption of water culture.

The self-watering method has more practical for water replacement in the soil in pots. Sunflower production was influenced by soil moisture maintenance methods in cultivation under controlled conditions.

Conflict of Interests

The authors have not declared any conflict of interests.

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Performance of eleven introduced improved lowland rice varieties in the northern Savanna zones of Ghana

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From 2007 to 2010, we evaluated eleven introduced improved rice materials, mainly lowland NERICAs and Thailand varieties for yield and other farmers' preferred agronomic traits compared to Gbewaa (aromatic local check) and Digang (non aromatic local check). The trials, each year, were laid out in a Randomized Complete Block Designed (RCBD) with four replicates at two locations (Nyankpala and Salaga) in Northern Region, Ghana. Six cultivars namely, WAS 163-B-5-3, WAS 122-13-WAS-10-WAR, L2-4, PERFUME IRRIGATED, LONG GRAIN ORDINARY 2 and EXBAICA yielded significantly higher ($p<0.5$) with mean yield advantage range of 21 to 73% over Gbewaa, 16 to 66% over Digang, adapted to the target environment and tolerant to the major biotic stresses. These materials include two aromatic varieties which could serve as substitutes for Gbewaa (the market aromatic quality) and give farmers a choice to meet the existing market demand for aromatic rice.

Key words: Rice evaluation, Savanna agro ecologies of Ghana, NERICAs, improved varieties.

INTRODUCTION

Rice has successfully become a staple food in Ghana. This is a result of the rapid change in lifestyle and food habit; particularly in urban centers (Nyanteng, 1987; Balasubramanian et al., 2007). The per capita rice consumption has risen from average of 17.5 kg in 2001 to 22.6 kg in 2004 and is estimated to reach 63 kg in 2015 (JICA, 2007).

Ghana continues to import about 60% of its rice needs and consequently spends about US$ 450 million annually on imports (MOFA, 2010; Mohapatra, 2012). Efforts to increase domestic rice production will contribute directly to foreign currency saving and balance of payment, in addition to improving farmers' incomes. Increased productivity from increased yield per unit area through the use of high yielding locally adapted varieties and good agronomic practices is the most viable strategy to reduce dependency on importation (MOFA, 2009).

Ghana mostly depends on the Consultative Group on International Agricultural Research (CGIAR) centers, that is, Africa Rice Centre (AfricaRice) and International Rice Research Institute (IRRI) for its improved breeding materials (MOFA, 2009).

The Participatory Varietal Selection approach (PVS), which aids easy access of promising materials to farmers (AfricaRice, 2010), is mostly employed in evaluating the materials. Promising ones tested at multi-locations and

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Table 1. Varieties and sources used for the Multi-location trials.

Variety	Source	Year introduced/released
WAS 122-IDSA-13-WAS-10-WAB-B-TGR5	AfricaRice	2004
WAS 163-B-5-3	AfricaRice	2004
L-2-4	AfricaRice	2004
WAS 122-IDSA-11-WAS-11-4-FKR1	AfricaRice	2004
WAS 161-B-6-4-FKR1	AfricaRice	2004
Thai 3(Long grain ordinary rice 2 irrigated)	Thailand	2008
Thai 4 (heavy jasmine lowland)	Thailand	2008
Thai2 (Long grain ordinary rice 1 irrigated)	Thailand	2008
Thai 1 (Perfume irrigated)	Thailand	2008
Thai 5 (Light Jasmine Lowland (95 days)	Thailand	2008
EXBAIKA	University Farm, Kpong	2004
DIGANG*	SARI	2002
GBEWAA RICE/ JASMINE 85*	USA	2002/2009**

*Released varieties used as local check; ** Introduced in 2002 and released as a variety in 2009.

on-farm are recommend for release as varieties.

The savanna zones of Ghana which constitute the three Northern regions (that is, Northern, Upper East, and Upper West) produce over 60% of the local rice (JICA, 2007). The Savanna Agricultural Research Institute (SARI), one of the 13 institutes of the Council for Scientific and Industrial Research (CSIR) is mandated to conduct research and introduce improved technologies to enhance agricultural productivity in this zone.

As part of its mandate, new higher yielding rice varieties are to be introduced to farmers to enable them improve their productivity. This paper presents the performance of introduced lowland NERICAs and some Thailand materials in the Savanna agro ecological zones of Ghana.

MATERIALS AND METHODS

From 2007 to 2010, we evaluated at Nyankpala and Salaga five lowland NERICA Varieties, 5 introductions from Thailand, one introduction from University of Ghana Research station (Kpong) and two local control varieties. The five Lowland NERICAs were the most preferred farmer selections out of 30 entries during a previous Participatory Variety Selection (PVS-R) in 2004 (SARI, 2010). The introductions from Thailand and Kpong were included in 2008. The list of varieties used for the multi-location tests are presented in Table 1.

Experimental sites

The materials were tested at CSIR-SARI main station at Nyankpala and sub-station at Salaga. The Nyankpala site which falls within the Guinea savanna agro ecological zone is located 16 km West of Tamale the capital city of northern region of Ghana. The area lies in latitude 9° 25' 41" N and longitude 0° 56' 42" W, altitude 183 m above mean sea level. Annual average rainfall is about 1000 mm and mean temperature of 39°C (Kasei, 1988). The Salaga site falls within the west savanna transitional agro-ecological zone and is located 5 km south of Salaga the capital city of East Gonja district. The area lies in latitude 8° 33' 25" N and longitude 0° 31' 04" W, 156 m above mean sea level. Annual average rainfall is 1100 mm and mean temperature 27.9°C (Kasei, 1988). Weather conditions during the experimental years at Nyankpala is presented in Table 2.

Characteristics of the experimental soil

The characteristics of the experimental soils at the two sites are presented on Table 3. Total N was determined by the Kjeldahl procedure (Tel and Hegatey, 1984), available P by the Bray 1 extraction solution procedure (Bray and Kurtz, 1945) and exchangeable K by 1.0 M neutral NH_4OAc solution (Black, 1965). According to the physical and chemical characteristics, the soils can be described as deep sandy loam to clay loam (Buri et al., 2012). The pH was 4.8 at the 0 to 15 cm level and 4.9 at 15 to 30 cm. Compared to Buri et al. (2012), the levels of N, P and K in the soils were considered to be low.

Field preparation and experimental design

The experimental sites each year were ploughed around the first week of June and harrowed a week or two after (at the middle of June). A Randomized Complete Block Design (RCBD) with four replicates and a unit plot size of 3 m x 5 m was used.

Planting and cultural practices

Planting was done by dibbling seeds in a space of 20 cm × 20 cm between rows and hills. One seedling was allowed per hill three weeks after sowing. The recommended rate of compound fertilizer N-P-K of 60-60-30 kg/ha was applied in two split applications (Ragasa et al., 2013). Three weeks after each planting date, the basal fertilizer of 30-60-30 kg/ha NPK (grade 15-15-15) (300 g) and triple superphosphate fertilizer (98.7 g) were applied per experimental plot of 15 m^2. The remaining nitrogen was applied as top dressing at seven weeks after planting using sulphate of ammonia (214 g/plot). Weeds were initially controlled with pre-emergence herbicide (Pendimethaline 400 g/l; Alligator) at 3.2 L/ha a day after planting and then by hand weeding. Rice was harvested and threshed manually at maturity using sickles and beating on tarpaulin respectively. After measurement of grain moisture, weight

Table 2. Summary of weather data for May to October at Nyankpala for the experimental years.

Expt year	Data	Rain fall (mm)	Rain days (mm)	Evaporation (kph)	Wind speed (km/hr)	Sun shine (°C)	Min temp (°C)	Max Temp (°C)	Mean Temp (%)	Min Rel-Hum (%)	Max Rel-Hum (%)	Mean Rel-Hum (%)
	Total	734.4	52	-	19.7	31.1	138.9	186.8	163	414	542	478
2006	Mean	122	8.7	-	3	7.3	22.7	33.8	28.2	55	79	67
	Total	873.5	54	624.14	20.6	39.4	141	188.5	164	419	543	481
2007	Mean	146	9.0	104	3	7	24	31	27	70	79	66
	Total	1184.9	72	738.25	20.13	58.79	269.8	404.1	336.8	680	949	819
2008	Mean	197	12	123	3	4.89	22.5	33.7	28.1	57	79	68
	Total	1209.1	53	717.34	16.25	14.6	140.8	202	172.3	437	554	497
2009	Mean	202	8.8	120	2.87	7.84	23.2	35.6	29.5	59	82	70

Table 3. Physical and chemical characteristics of soils used.

Site	% Sand	% Silt	% Clay	pH (H_2O)	% OC	% N	P (mg/kg)	K (mg/kg)
Nyankpala	57.8	40.2	2.0	4.8	0.57	0.046	10.75	49.02
Salaga	53.96	21.72	24.32	4.5	1.07	0.088	8.65	48.63

per plot yield of winnowed paddy was assessed at 14% moisture content.

Traits evaluation

Traits evaluated for the study include grain yield, plant height, number of tillers, number of panicles, days to 50% flowering, maturity days, 1000 grain weight, disease scores for blast, brown spot and leaf scald based on IRRI's Standard Evaluation System for rice (IRRI, 1996).

Statistical analysis

Analysis of variance was first performed (SAS Statistical Package) followed by LSD for mean comparison and separation when necessary.

RESULTS

Results of selected parameters measured in 2007 are summarized in Table 4. The recorded yields for the two locations; Nyankpala and Salaga in 2007 are also presented in Table 5.

The yield performance for the years 2008, 2009 and 2010 are presented in Table 7.

DISCUSSION

Generally, the recorded yields were low compared to previous report (JICA, 2007). The yield ranged between 0.7 and 5.5 t/ha for Nyanpkala and 0.8 to 4.5 t/ha at Salaga over the years. Gbewaa, the aromatic check recorded average yield of 2.28 t/ha (Table 7) over the years which is just about 40% of its potential yield of 6 to 7 t/ha (SARI, 2008). Intermittent moisture stress could account for the recorded low yields since experimental fields were rain fed and the plants occasionally suffered from severe moisture stress during the various seasons especially at Salaga in 2010 where the recorded yields were particularly low (Table 7). Terminal season drought is the major abiotic constraint of rice production in the savanna regions (MOFA, 2009). Digang, the non aromatic check averagely performed better (2.4 t/ha) than Gbewaa (2.28 t/ha) (Table 7).

Digang is known to tolerate drought better than Gbewaa. Gbewaa is reported to have problem

Table 4. Plant parameters measured in 2007.

Genotype	Average plant height (cm)	Maturity (days)	Average number of tillers/hill	Disease scores		
				L. Blast	B. Spots	L. Scald
WAS 122-13-WAS-10-WAB-B-TGR5	126	126	14	1	2	3
WAB 163-B-5-3	115	126	12	1	2	3
L2-4	106	111	13	1	3	1
WAS 122-IDSA-10-WAS 11-4-FKR1	93	115	16	0	1	0
WAS 161-B-6-4-FKRI	99	112	16	1	0	0
DIGANG	118	113	9	1	1	0
JASMINE 85	101	116	16	0	1	3

Table 5. Yield (t/ha) of promising varieties at the two sites in 2007.

Variety	Nyankpala	Salaga	Mean
WAS 122-13-WAS-10-WAB-B-TGR5	5.3	4.3	4.8
WAB 163-B-5-3	5.5	4.5	5.0
L2-4	5.0	4.4	4.7
WAS 122-IDSA-10-WAS 11-4-FKR1	5.0	4.4	4.7
WAS 161-B-6-4-FKRI	4.4	4.1	4.3
DIGANG	4.5	4.0	4.3
JASMINE 85	4.3	4.0	4.2
LSD (0.05)	0.6	0.2	-

with drought and diseases (Acheampong, 2010). Planting early and medium duration varieties has been the reliable strategy to escape the effect of terminal season droughts. The selected materials are of medium duration (ranging from 108 to 130 days) (Table 6) which makes them suitable for cultivation in the region. However, to ensure stable yields and sustainable cultivation of these materials in the drought-prone savanna zones, effort should be made to improve the level of drought tolerance of these materials. Analysis of variance across the two locations showed no significant genotype by location interaction indicating that the selected materials can be grown across the target environment.

The selected materials have plant height range of 96 to 126 cm, (Table 6), mostly semi dwarfs to avoid lodging. The materials also have adequate tolerance to brown spot, leaf blast and leaf scald (Table 6); the most prevalent diseases in the tested environment. Screening against these diseases was incorporated in the evaluation process right from the observational nursery scoring each year based on the Standard Evaluation System (SES) of rice by IRRI (1996). The materials also have appreciable levels of tolerance to African gall midge; the most prevalent insect pest (Table 6).

The five introductions from Thailand were included in 2008 because of the market demand for aromatic rice. The Ghanaian market has high preference for intermediate amylose, long slender aromatic rice grains, with these characters accounting for over 40% increase in price quotations (Asante, 2005; Minkah, 2007; Anang et al., 2011). Gbewaa rice is cultivated by majority of Ghanaian farmers because of its aromatic quality. However, it is susceptible to blast and Rice Yellow Mottled Virus (RYMV) (Asante, 2012). Digang (the non aromatic check) is resistant to these two diseases but not preferred because of its poor grain quality (Asante, 2012).

Table 6. Agronomic characteristic of Lowland NERICAs and introduced varieties from Thailand in 2008.

Variety	No. of tillers/hill	Days at 50% flowering	Days to maturity	Plant height (cm)	No. of panicles per hill	Brown spot	Leaf blast	Leaf scald	Gall midge
L2 - 4	13.9	89	118	95	9.3	2.25	0.5	3	0
WAS 161-B-6-4-FKR1	14.3	93	117	92.9	9.7	1	0	3.5	3
WAS 163-B-5-3	12.7	89	117	105.8	9	3	0.25	3.5	0
Thai 3 (long grain ordinary rice2 irrigated	10.3	98	130	124.4	7.7	1.25	0.25	3	3
WAS 122-13-WAS-10-WAB-B-TGR5	12	93	126	101.4	9.1	4	0.5	3	0
Jasmine 85	13.6	89	116	93.9	7.8	3	0	3	3.5
Thai 4	13.8	114	133	143.5	4.8	1.5	0	4.5	4.5
Digang	12.2	87	113	108.8	6.8	1.75	0.5	2.5	0
WAS 122-IDSA-11-WAS 11-4-FKR1	14.1	90	117	86.5	9.6	1.25	0	6	6
Exbaika	12.7	98	122	90	9.2	3.25	0.25	4	5.5
Thai 2	11.7	77	108	92.3	9.4	2	0.25	3	0
Thai 1 (Perfume irrigated (110 days)	14.2	110	131	104	7.1	1	0	6.5	6.5
Thai 5	11.2	106	129	137.7	7.2	1.5	0	4	7
Lsd(0.05)	2.1	7.8	5.5	8.4	1.81	1.33	NS	1.19	1.1

Diseased scores based on IRRI's SES (1996).

Table 7. Yield (t/ha) of varieties at the two sites in 2008, 2009, and 2010.

Variety	Yield (t/ha)					Mean & SD
	2008	2009		2010		
	Nyank	Nyank	Sala	Nyank	Sala	
L2 - 4	3.9	4.5	3.3	4.3	3.9	3.98 ± 0.632
WAS 161-B-6-4-FKR1	3.7	3.0	1.5	2.4	1.0	2.32 ± 1.094
WAS 163-B-5-3	3.6	4.2	2.8	4.1	3.7	3.68 ± 0.554
Thai3 (Long grain ordinary rice 2 irrigated)	3.2	4.0	3.4	3.4	3.0	3.4 ± 0.374
WAS 122-13-WAS-10-WAB-B-TGR5	3.2	3.8	3.0	4.1	3.9	3.6 ± 0.474
GBEWAA (L/C)	2.9	3.0	2.0	2.4	1.1	2.28 ± 0.773
Thai 4 (heavy jasmine lowland)	2.8	2.5	2.0	0.7	0.9	1.78 ± 0.942
DIGANG(L/C)	2.7	2.9	2.5	2.5	1.4	2.4 ± 0.583
WAS 122-IDSA-11-WAS 11-4-FKR1	2.6	3.6	1.8	3.0	1.6	2.52 ± 0.832
EXBAIKA	2.6	3.8	3.0	4.1	3.9	3.48 ± 0.646
Thai 2 (Long grain ordinary rice 1 irrigated)	2.5	2.1	1.1	2.1	0.8	1.72 ± 0.729
Thai 1 (Perfume irrigated)	2.2	3.0	2.6	1.8	1.4	2.2 ± 0.632
Thai 5 (Light Jasmine Lowland (95 days)	2.0	2.8	2.3	2.5	1.0	2.12 ± 0.691
Lsd (0.05)	0.8	0.4	0.3	0.8	0.2	-

Nyank=Nyankpala, Sala=Salaga, SD=Standard deviation.

Two of the Thailand introductions (PERFUME IRRIGATED and LONG GRAIN ORDINARY 2) proved suitable for cultivation in the savanna ecology. Other interesting characteristics of the selected materials include high tillering ability, large panicle size and long grains preferred by the Ghanaian market.

Conclusion

Six of the test materials (WAS 163-B-5-3, WAS 122-13-WAS-10-WAR, L2-4, PERFUME IRRIGATED, LONG GRAIN ORDINARY 2 and EXBAICA) outperformed the local popular checks (Digang and Gbewaa) and are currently undergoing on-farm testing pending possible recommendation for release as varieties. Two aromatic varieties (PERFUME IRRIGATED and LONG GRAIN ORDINARY 2) could serve as substitute for Gbewaa and give farmers a choice to meet existing demand for aromatic rice.

Conflict of interest

The authors have not declared any conflict of interest

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Genetic variability of Ethiopian bread wheat genotypes (*Triticum aestivum* L.) using agro-morphological traits and their gliadin content

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Wheat is grown worldwide because it is a major diet component. The aim of this study was to investigate the genetic variation in bread wheat (*Triticum aestivum* L.) when grown in the Wollo highland areas, applying agro-morphological data and gliadin content. A total of twenty accessions of bread wheat received from Ethiopian Biodiversity Institute (EBI), Sirinka Agricultural Research Centre (SARC) and also newly collected accessions were planted in a randomized complete block design, starting mid-January, 2014, using irrigation. Agro-morphological data was recorded following the International Plant Genetic Resources Institute (IPGR) descriptors list, and the gliadin content was examined by acid page electrophoresis. Analysis of variance showed that most agro-morphological data varied significantly between accessions. By using both dendrogram based on agro-morphological traits and cluster analyses of gliadin, the accession 226944 and the landrace Debalit were found to cluster closely. Newly collected accessions and EBI accessions showed higher diversity than released varieties applying Nei's genetic distance and gene diversity (H*) analysis. Accessions originating from the West part of the South Wollo province were found to be grouped together, applying principal component analysis. It can be concluded that high genetic variability exists on the bread wheat genotypes, some of them holding exceptionally promising values, making them interesting for further selection.

Key words: Acid page electrophoresis, agro morphological traits, Ethiopia, genetic variability, gliadin, *Triticum aestivum*.

INTRODUCTION

Wheat is grown on more land area worldwide than any other crop and is rated third to rice and corn in total world

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production (Western Organization of Resource Councils (WORC), 2002). Wheat is grown on more than 240 million ha and the world trade is greater than for all other crops combined. Wheat is a major diet component due to the wheat plant's agronomic adaptability, ease of grain storage and ease of converting grain into flour for making edible, palatable, interesting and satisfying food (Food and Agricultural Organization (FAO), 2002; Asif et al., 2005). Considering all the *Triticum* species (as will be seen later), it is a major food crop and supporting 35% of the world population (Schuster et al., 2009; Sehgal et al., 2012). In Ethiopia, the crop ranks third in terms of total production, next to teff and maize (Hailegiorgis et al., 2011).

Ethiopian wheat belongs to the following species: *Triticum (T) diccoccum, T. turgidum, T. polonicum, T. durum, T. aestivum, T. compactrim* and *T. abyssinicum.* Nowadays, most of the wheat grown in Ethiopia is bread wheat (*T. aestivum* (Central Statistics Agency (CSA), 2013). Ethiopia is the second largest wheat producer in sub-Saharan Africa after South Africa. Most parts of Ethiopia are wheat producing, however, the most suitable area falls between 1900 to 2700 m.a.s.l (Hailu et al., 2006). Wheat is mainly grown in the highlands, planted in the summer before the main rainy season, and harvested in October to November. Seventy-five percent of the wheat is grown in Arsi, Bale, Shoa and Wollo provinces (CSA, 2013). Wheat production in Ethiopia in the 2012 to 2013 season accounted for 3,434,706 tons, with 888,569 tons in Amhara region and 130,595 tons in south Wollo zone specifically (CSA, 2013). Although Ethiopia has seen relatively steady improvement in wheat production in recent years, both in the amount of arable land cultivated and the per hectare yields, the growth has been a fraction of what it could be with more focused extension efforts in place (ATA, 2013), including a better understanding of impact of water resources (Valipour 2012; 2014a, b, c, d, e, 2015a, b, c, d; Valipour et al., 2015).

Wheat storage proteins, namely gliadins and glutenins, are the main components of gluten, which is the main contributor to the rheological and bread-making properties of wheat (Branlard et al., 2001; Johansson et al., 2013). Glutenin subunits are classified as high molecular weight (HMW) and low molecular weight (LMW) subunits on the basis of their mobility in sodium deodocyle sulphate poly acrylamide gel electrophoresis (SDS-PAGE). Glutenins confer elasticity to dough, whereas gliadins are viscous and give extensibility to dough (Payne et al., 1984).

Gliadins are heterogeneous mixtures of single-chained polypeptides with a molecular weight range of 30,000 to 75,000 Da. Due to extensive polymorphism, these proteins have been widely used for cultivar identification in hexaploid and tetraploid wheat's (Payne et al., 1984). Allelic variants differ in the number, mobility, and intensity of their components and can be characterized through acid poly acrylamide gel electrophoresis (A-PAGE) (Tatham and Shewry, 1985) or SDS-PAGE (Damania et al., 1983).

Various researchers from Ethiopia have made investigations on genetic diversity of wheat, applying molecular (Tsegaye and Tessema, 1995; Hailu et al., 2005), agro morphology (Tessema et al., 1991; Bechere et al., 1996; Hailu et al., 2006; Hailegiorgis et al., 2011) and protein quality (Dessalegn et al., 2011) methods. Quantitative diversity analysis of wheat shows that the coefficient of variation differs between regions both for tetraploid and hexaploid wheat, thereby being an indication of different levels of variation within the region (Bekele, 1984). Most of the studies have focussed on analysing the genetic distance between few varieties of samples collected from the central part of the country mainly/only.

The aim of this study was to evaluate genetic variation in various wheat materials originating from different areas of Ethiopia by growing them in the South Wollo province. We had the hypothesis that genetic variation analysis may vary in relation to the areas the field trials are carried out. The present wheat material has not been evaluated previously in the Wollo province. Furthermore, suitable genotypes for a certain environment have to be evaluated in the same environment as most characters are not only influenced by the genotype but also of the environment in which they are grown. Three landraces originating from the Wollo province, and not evaluated before, were included in the study. In order to study genetic variation originating from the specific cultivation regions with genetic variation not being influenced by the environment, both agro-morphological traits and specific protein composition analysis were carried out to study the genetic variation.

MATERIALS AND METHODS

Plant materials and experimental site

A total of twenty genotypes of bread wheat (*Triticum aestivum* L.), that consisted of ten accession obtained from the Ethiopian Biodiversity Institute (222815, 212648, 222853, 222680, 243696, 243697, 243704, 7506, 7565, 226944), three landraces collected by Dimiru Tilahun from the South Wollo administrative province (namely: Jiru, Tikur Sinde, and Debalit), and seven released varieties (Dinknesh, ET-13A, Menze, Sora, Tsehay, Tossa, Warkaye) obtained from Sirinka Agricultural Research Centre (SARC), were used for this study. The field experiment to collect the data on agro-morphology and seeds for gliadin analysis was carried out at the Wollo University Dessie campus. The site is located at 11° 10' 10'' North and 38° 39' 16'' East, and at an elevation of 2475 m above sea level, 406 km north of Addis Ababa. The area receives annual rainfall ranging from 900 to 1400 mm and temperature range from 12.5 to 27°C. The analysis of gliadin composition was carried out at Ethiopian Biodiversity Institute (EBI) laboratory at Addis Ababa on July, 2014.

Table 1. Descriptive statstics of the traits based on genotypes used for the study.

Traits	Range	Minimum	Maximum	Mean	Standard error	Standard deviation	Variance	Skewness
Days of heading (no)	16.00	62.00	78.00	68.18	0.66	5.13	26.32	0.48
Days of maturity (no)	17.00	115.0	132.00	122.53	0.62	4.79	22.97	0.82
Tiller number (no)	9.63	4.00	13.63	7.89	0.29	2.21	4.88	0.62
Plant height (cm)	54.88	55.88	110.75	76.54	1.44	11.13	123.84	0.67
Spike length (mm)	4.13	4.13	8.25	6.63	0.12	0.96	0.91	-0.51
Thousand kernel weight (g)	30.51	20.11	50.63	34.65	0.95	7.40	54.71	0.03
Grains per spikelet (no)	1.00	2.00	3.00	2.32	0.06	0.47	0.22	0.81
Grain yield (g)	667.6	86.56	754.14	343.41	23.15	179.33	32157.48	0.56
Biomass yield (kg)	3.00	1.00	4.00	1.42	0.09	0.73	0.53	1.13
Grains per spike (no)	25.00	17.50	42.50	29.29	0.75	5.85	34.19	0.11
Spike weight per plant (g)	2.19	0.68	2.86	1.55	0.06	0.47	0.22	0.31
Number of spikes per plant (no)	9.25	3.00	12.25	6.82	0.25	1.95	3.79	0.52
Grain weight per spike (g)	1.53	0.44	1.96	1.04	0.05	0.39	0.15	0.40

Experimental design

The field experiment was laid out in a randomized complete block design with three replications starting from January and ending in July, 2014. The plots sizes were 2 m × 1.2 m with a row distance of 20 cm and between plants 5 cm in six rows. Data was taken from the middle four rows and used for the analysis. Fertilizer was applied as per the recommendation of the Ministry/Bureau of Agriculture (150 kg DAP and 150 kg UREA ha^{-1}) (Ministry of Agriculture and Rural Development (MoARD), 2012).

Data collection

The data was recorded at maturity and grain yield was measured after measuring moisture content. In order to allow us to see fine details of changes, all the data were transformed to $\log_{10}$ before analysis.

Agro-morphological data

Observations on yield and yield attributing characters were recorded. In each plot, eight random plants were tagged to record these observations and mean values of the recorded values were calculated. The descriptions of characters studied was based on International Plant Genetic Resources Institute (IPGR) (1985) descriptors list that include days to heading (DH), days to maturity (DM), number of tillers (NT), plant height (PH), spike length (SPL), number of spikelet's per spike (NSPSP), number of grains per spike (GNPS), thousand kernel weight (TKWT), grain yield (GYPP), biomass yield per plot (BMYPP), presence or absence of Awn (AW), and grain number per spike (GNPSP).

Storage protein methodology - gliadin

The assessment of storage protein composition was carried out according to Tatham and Shewry (1985) with some modifications. Twenty four spikes from the randomly tagged plants of each accession were taken and threshed independently and conserved in a seed bags. After taking to EBI, A-PAGE was used to examine the composition of gliadin content based on the mobility of gliadin bands of wheat genotypes used for the study and bands were scored as present (1) or absent (0).

Data analysis

Analysis of variances (ANOVA), dendrogram (cluster) and principal component analysis (PCA) were applied on the agro-morphological data using the SAS software program (SAS, 2004). Nei's (1973) genetic diversity and cluster analysis were carried out using POPGEN version 1.31 and TFPGA version 1.3, respectively, based on gliadin bands composition.

RESULTS AND DISCUSSION

Agro-morphological variation

The genotypes showed variation based on agro-morphological traits (Table 1). For example, plant height averaged 76.53 cm and ranged from 55.9

Table 2. Analysis of Variance of the genotypes used in the study.

HI	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Block	2	0.004298	0.0021489	0.6913	0.507092
Genotypes	19	0.145512	0.0076585	2.4639	0.008847 **
Error	38	0.118117	0.0031083	-	-
DH					
Block	2	60.43	30.217	5.9939	0.005463 **
Genotypes	19	1300.98	68.473	13.5826	1.503×10^{-11} ***
Error	38	191.57	5.041	-	-
DM					
Block	2	47.43	23.717	2.9207	0.06608
Genotypes	19	998.93	52.575	6.4747	5.599×10^{-07} ***
Error	38	308.57	8.120	-	-
NT					
Block	2	66.133	33.067	8.7932	0.0007269 ***
Genotypes	19	78.565	4.135	1.0996	0.3888386
Error	38	142.898	3.760	-	-
GNPS					
Block	2	106.85	53.424	5.9055	0.005844 **
Genotypes	19	1566.36	82.440	9.1129	5.69×10^{-09} ***
Error	38	343.77	9.047	-	-
TKWT					
Block	2	362.69	181.345	8.5953	0.0008326 ***
Genotypes	19	2063.60	108.610	5.1478	8.941×10^{-06} ***
Error	38	801.73	21.098	-	-
GYPP					
Block	2	218505	109253	9.4857	0.0004554 ***
Genotypes	19	1241115	65322	5.6715	2.866×10^{-06} ***
Error	38	437671	11518	-	-
BMYPP					
Block	2	2.6249	1.31245	5.3975	0.008645 **
Genotypes	19	19.1829	1.00963	4.1521	9.311×10^{-05} ***
Error	38	9.2401	0.24316	-	-

Df= degrees of freedom; Sum Sq= Sum of Square; Mean Sq = Mean sum of square; Significant at $P < 0.001$,***; $P < 0.01$, **; $P < 0.05$, *HI (Harvest Index), DH (Days to Heading), DM (Days to Maturity), NT (Number of tillers), GNPS (Numbers of grans per spike), TKWT (Thousand Kernel Weight), GYPP (Grain Yiled) and BMYPP (Biomass Yield Per Plot).

to 110.8 cm; while days to heading averaged 68.18, with a range of 62 to 78. Significant variation was found both among genotypes and blocks for all characters except tiller number for genotype, days to harvest, and plant height for blocks applying ANOVA (Table 2).

Similar to the findings in the present study of genetic variation in agro-morphological traits, previous studies have shown genetic variation in grain number per spike and grain weight per spike (Zecevic et al., 2010), plant height, number of productive tillers per plant, number of spikelet's per spike, spike length, number of grains per spike, fertility percent, thousand kernel weight and yield

per plant (Ali et al., 2008). The present study showed correlation between yield and several of the morphological traits known to be components determining the yield, including number of grains per spikelet, number of tillers, biomass, spike weight, harvest index, number of seed per spikelet, disease score and number of grains per spike (result not shown). A previous study has shown correlation between yield and 19 other morphological traits (Ojaghi and Akhundova, 2010). It is well known that yield is a polygenic trait and that such traits are related to several other interacting traits. Benefits can be acquired by the use of other traits that are strongly correlated with the yield, indirectly increasing the yield (Johnson and Wichern, 2002; Hailu et al., 2005). Morphological traits such as plant height, plant development, maturation time etc have been used to determine the genetic distance between and within varieties (Hailu et al., 2006, 2010). Morphological traits have been used for selection of suitable plants since ancient times, although lately a combination with molecular markers is more common (Hailu et al., 2005). Morphological markers are inexpensive and simple to score as they are based on distinct phenotypes such as plant colour, plant height, seed characteristics, etc. but the main disadvantage of such markers is that in studies of genetic diversity, the expression of the phenotypes is highly influenced by environmental conditions (Ahmadi et al., 2012).

In the present study, we combined the morphological data with composition of gliadins in the material. Gliadin composition is a character that does not vary over environments (Huebner and Bietz, 1988) and it is a stable genetic character that can be used for analysis of genetic variation. Futhermore, protein composition in wheat materials are of large relevance as they determine baking and pasta making quality attributes in wheat (Johansson et al., 2005, 2008, 2013). The knowledge about the genetic relationships of genotypes provides useful information to address in breeding programs and germplasm resource management (Aharizad et al., 2012). Furthermore, an understanding of water resource requirements in relation to agronomic properties plays a role for the variation (Valipour, 2013).

Cluster analysis using agro morphological traits

Cluster diagram based on Euclidean dissimilarity using complete linkage method, at the rescaled distance of 10, categorized the genotypes into two major groups (Figure 1) whereas 222815 and 243697 were left solitary (ungrouped with the other accessions). Cluster one contains two genotypes, Debalit and 226944, while cluster two contain 16 genotypes. If rescaled distance instead is selected to be 5, the large cluster consisted of 14 genotypes, while all other genotypes were left solitary (Figure 1). Genetic diversity using cluster mean analysis by Euclidean dissimilarity dendrogram has also been reported previously in other studies (Ali et al., 2008; Shahryari et al., 2011; Aharizad et al., 2012; Ahmadi et al., 2012; Degwoine and Alamrew, 2013;). The dendrogram elaborate the relative magnitude of resemblance among the genotypes as well as the clusters (Singh et al., 2014).

The present result showed that all released varieties except Workaye were grouped in the large group of 14 genotypes that were grouped at the rescaled distance of 5. The landraces/accessions obtained from EBI did not group closer with each other than they did with the released varieties, indicating that the landraces were not closely genetically related than their genotypic relationships were with the varieties. The accession 222815 was the one most distantly related to the other genotypes.

Storage protein analysis

The highest Nei's gene diversity, 0.295, was observed in accession 222815, one of the landraces obtained from EBI, while the lowest value was found in the released variety ET-13A (0.00). Both the accessions obtained from EBI and the three local varieties showed comparatively higher Nei's gene diversity compared to the released varieties (Table 3). Thus, similarly as has been found in previous studies (Hailu et al., 2010), the general highest gene diversity was found among landraces while the lowest gene diversity was found among released varieties in the present study, which might be expected as variety development by breeding is supposed to result in a pure variety. However, even bred varieties can sometimes lack uniformity (Husenov et al., 2015).

In the present study, the percentage of polymorphic bands in the accessions ranged from 21.74% (222680) to 82.61% (222815); for the new collections from 47.83% (Debalit and Tikur sinde) to 69.57% (Jiru). The released varieties showed the lowest percentage of polymorphic bands from 0.00% (ET-13A) to 21.74% (Dinkinesh and Warkaye). Metakovsky and Branlard (1998) reported that genetic diversity in genotypes can be determined by electrophoretic patterns of gliadin in the genotypes. Variation of wheat proteins is also used for determination of gluten strength and bread-making quality (Malik et al., 2013). Determination of gliadin is therefore useful not only in genotype identification but also in breeding programs (Bushuk and Zillman, 1978). Specific gliadin composition of genotypes is free from environmental factors making this an easy and convenient method in evaluation of germplasm variability, for pure seed productions in hexaploid wheat (Murat et al., 2013). Thus one of the reasons to use variation in gliadin composition for evaluation of genetic diversity is that these proteins

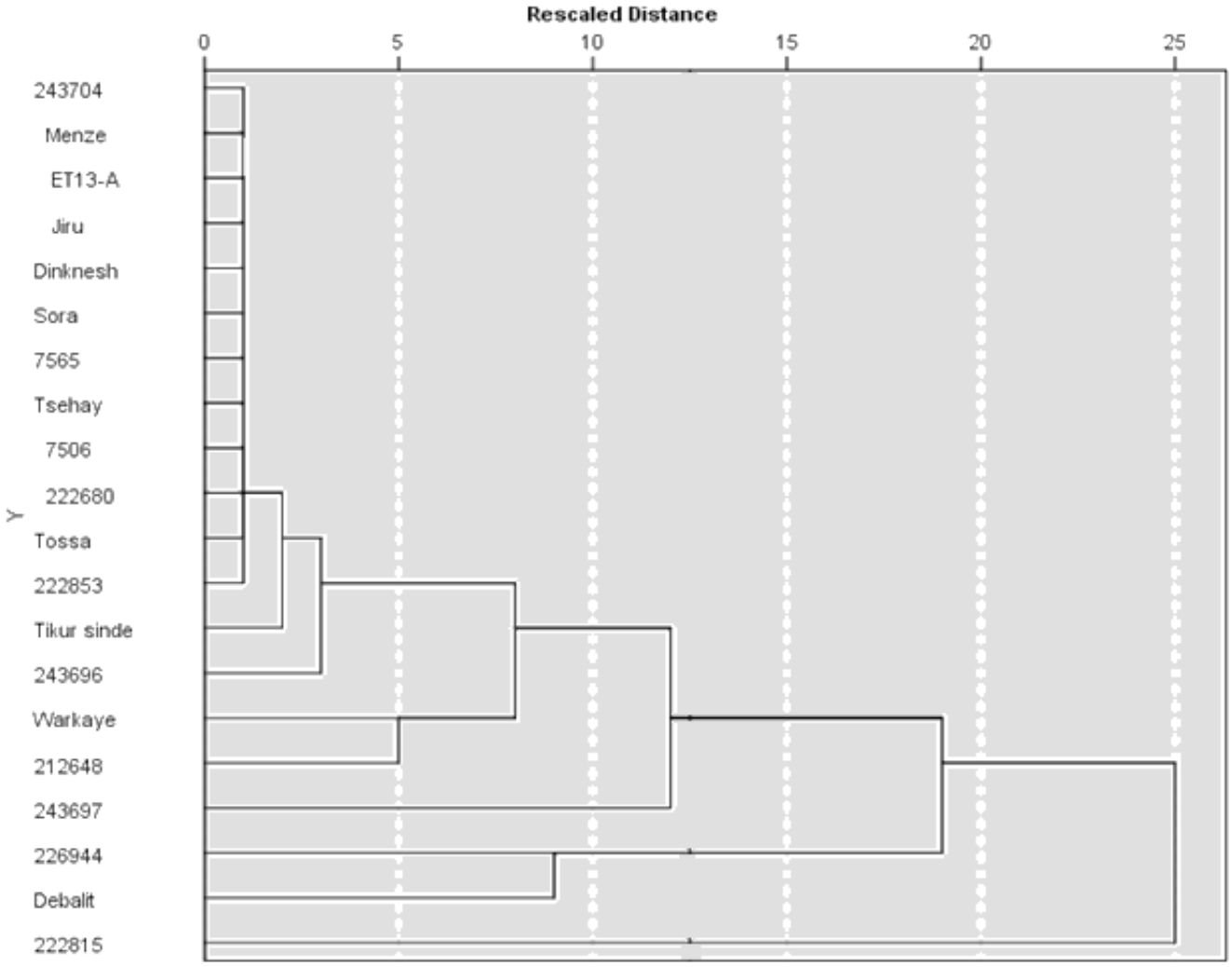

Figure 1. Dendrogram using average linkage (within groups) of the genotypes based on agro-morphological data.

are highly polymorphic, while their composition is not influenced by the environment but only by genetic variation (Huebner and Bietz, 1988). The information from gliadin analyses can also be combined for a better understanding of bread-making quality in the material (Johansson et al., 2013). Thus, their analyses in the present paper will help to identify genetically determined variation and also to compare if similar variation is obtained with agro-morphological data, the later also affected by the environment (Fufa et al., 2005; Ali et al., 2013).

Cluster and PCA analysis based on gliadin composition

By following the method by Zarkti et al. (2010), setting the distance for clustering to 2.5, the cluster analysis using gliadin showed that sample 222680, Tsehay, Menz and sample 243696 were left solitary (Figure 2). The other genotypes were clustered into three clusters. Debalit, 222853 and 226944 were clustered in cluster one; Dinknesh and Sora were clustered in cluster two; and the rest grouped together in cluster three, although clustering in three clusters at the distance for clustering of 1.50. While comparing the dendrogram based on agro-morphological traits with the cluster from cluster analyses of gliadins some similarities were recorded (Figures 1 and 3). One accession in both analyses was found to be the most distantly related genotype. Furthermore, the accession 226944 and the landrace Debalit were found to cluster closely in both analyses.

Principal component analysis (Figure 3) based on gliadin composition showed accession number 222853,

Table 3. Nei's gene diversity and percentage of polymorphic bands according to Nei, 1973.

Sample	Accession	Gene diversity (h*)	Percentage of polymorphic bands
1	222815	0.295	82.61
2	222680	0.056	21.74
3	7565	0.193	60.87
4	243704	0.182	60.87
5	243697	0.182	52.17
6	222853	0.121	47.83
7	243696	0.174	56.52
8	226944	0.169	56.52
9	212648	0.213	65.22
10	7506	0.251	73.91
11	Debalit	0.119	47.83
12	Tikur sinde	0.161	47.83
13	Jiru	0.180	69.57
14	Sora	0.029	8.70
15	Dinkinesh	0.07	21.74
16	Menze	0.039	8.70
17	Warkaye	0.088	21.74
18	Tsehay	0.029	8.70
19	Tossa	0.039	13.04
20	ET-13 A	0.00	0.00

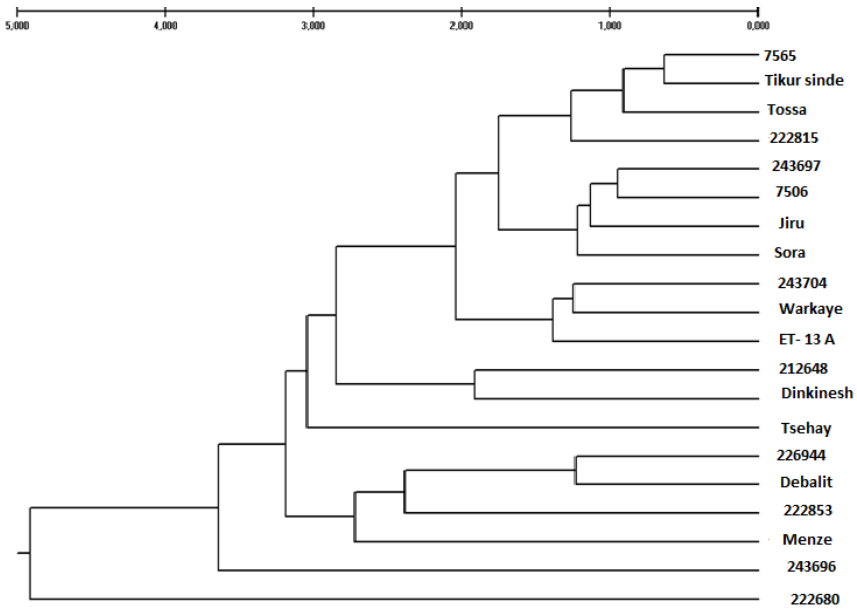

Figure 2. Dendrogram showing the linkage of the genotypes using gliadin.

243697, and the landraces Jiru and Tikur sinde to be the most different compared to the other genotypes analysed, the rest were mainly grouped into a large group (Figure 3). Passport data of the four mentioned genotypes showed that they are all collected from the Western part of the South Wollo administrative province. One nice idea would have been to make a more thorough analysis of the correlation between genetic distance

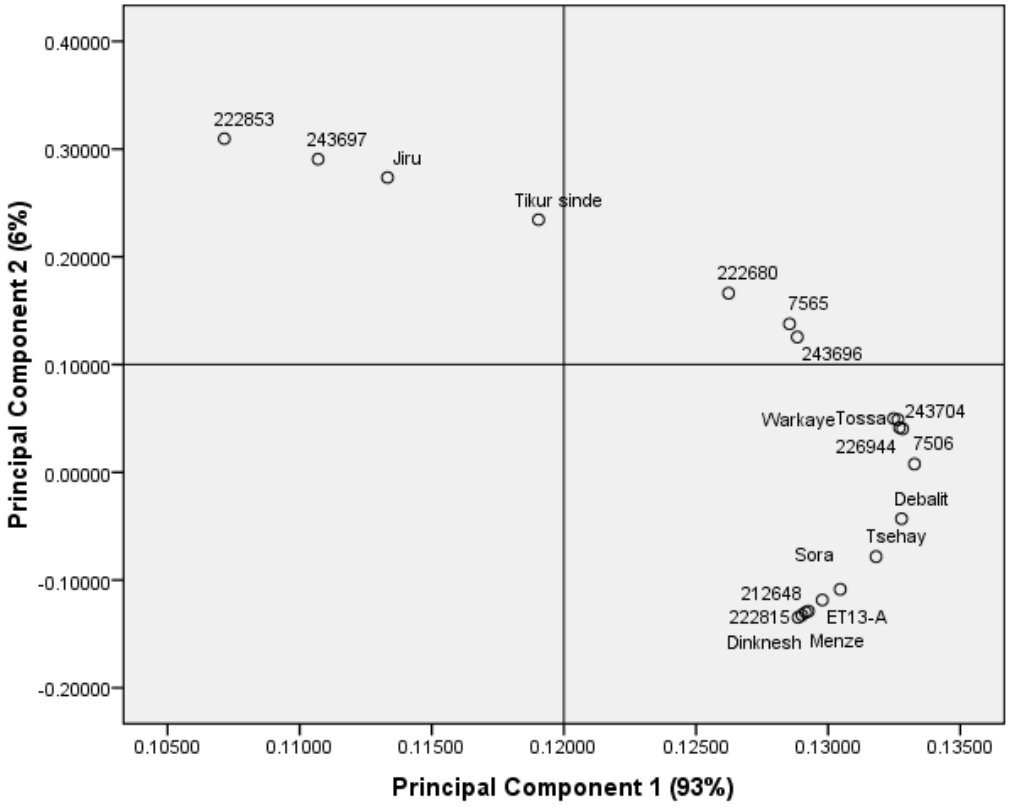

Figure 3. Biplot of Principal Components PC1 vs PC2 of Ethiopian bread wheat accessions.

among the genotypes and the geographical distribution of the genotypes as to distance separating their origin. However, the passport data of the genotypes did not allow such a comparison. Results in both cluster and principal component analysis showed the presence of variations between germplasm especially among accessions, although the same genotypes were not obviously the most distantly related ones with the different analyses. Khodadadi et al. (2011) states that cluster analysis based on principal component analysis is a more precise indicator of differences among wheat genotypes than cluster analysis not based on principal component analysis. In the present study used cluster analysis based on principal component analysis to be able to clearly see differences among genotypes.

Conclusion

This study on bread wheat showed that there was a significant variation between genotypes from EBI (accessions), SARC (released) and new collections using both agro-morphological and gliadin analysis. Different methods applied indicated various genotypes to be most distantly related as compared to the other genotypes. However, the study was able to reveal presence of genetic variation, both including and excluding environmental interactions in the investigated wheat material. Thus, the investigated wheat can thereby be utilized as sources to improve the bread wheat at country wide level thereby helping to alleviate poverty in the country for the future.

Conflict of Interests

The authors have not declared any conflict of interests.

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Full Length Research Paper

Break dormancy, germination capacity of medics after different techniques of scarification (Physical, Chemical and Mechanical)

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Medics are annual forages very interesting in pastures of Mediterranean and steppes rangelands. Such as other leguminous, they are subject to dormancy which delay and reduce germination. The response of these species (*Medicago polymorpha, Medicago intertexta, Medicago ciliaris, Medicago truncatula, Medicago muricoleptis*) to mechanical, physical and chemical scarification, applied for break dormancy, had been studied. Germination is almost full (reaching 100%) is fast (24 h) after scarification with sand-paper, which demonstrates that dormancy is exclusively imposed by teguments of the grain. Soaking in boiled water (20 s by population), passing through sulphuric acid during 5 min and 180 mn, passing in freezer and in refrigerator (respectively 2 h and 5 days), in drying oven at 115°C during 20 mn eliminates partially dormancy. However, passing through ovine digestive tract followed by germination test possesses the same effect as the witness. In exposure in ultra-low temperature at 196°C (liquid nitrogen) during 25 mn improves germination and grains germination speed to all quoted species which is comparable to scarified grains results by sand-paper.

Keys words: Scarification, medics, populations, germination, dormancy.

INTRODUCTION

Fertilisation is a result of seed, in flowers plants, angiosperms, the fertilisation is double. The grain is constituted of several different types of origin tissues: embryo, albumen and teguments.

Embryo and albumen are tissues of fertilisation. At the periphery of the grain, we find teguments, protecting sleeves being more or less resistant. They are maternal origin and drift from ovary tissues (Brunel, 2008). These organs are intended to give birth to a new generation of plants; during germination process they ensure the survival of species. They are adapted organs to resist in adverse conditions. Such resistance allows them to reach, often for a very long time, in substantially inert state, conditions that will allow them to germinate (Mullon and David, 1982). These transient aptitudes to the germination typically called dormancy.

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Table 1. Code and origins of studied populations and so average weight of 1000 grains.

Species	Populations code	Weight of de 1000 grains	Origins
M. truncatula	Tr201 Tr55	3.58 g	Algeria (2004 and 2012)
M .intertexta	I756 I52	15.77 g	Algeria (2004 and 2012)
M.ciliaris	S7 S5	13.21 g	Algeria (2004 and 2012)
M .polymorpha	poly 60 Poly 205	3.24 g	Algeria (2004 and 2012)
M. muricoleptis	Aus106	6.26 g	Turkey (2004 and2012)

We can consider it as an exceptional resistance of seeds to unfavourable conditions in exterior environment but also, and above all a very high level of dehydration state of the seed. Furthermore, desiccation seems to be more active than grain's drying. It is an active phase in terms of gene and metabolism expression. Which implied that preparation of grains for germination is already starting during desiccation of seeds (Angelovici et al., 2010). According to Pinfield and Dungey (1985) a lot of time and efforts had been made to try to understand and the difference between regulation mechanisms of maintenance dormancy, perception of inductive conditions, the break dormancy and the germination process. Mayer and Poljakoff-Mayber (1982) explained; dormancy may be on account of the embryo's immaturity, imperviousness of seed coat to water or to gas, preventing of embryo development because of mechanical reasons, of particular requirements of temperature or of light, or presence of inhibitory substances of the germination. Others authors attribute dormancy to physiological properties and / or morphological of the seed. According to Baskin and Baskin (2004), it may be classified in five large categories: physiological, physical, morphological, morpho-physiological and combinatory (physical and physiological). Generally, physical dormancy is caused by one or several waterproof layers of palisades cells macrosclerides in tegument (Baskin and Baskin, 2004).
However observations by scanning electron microscope revealed a thicker layer of palisades cells in tegument of *Medicago orbiculairis* compared to *Astragalus hamosus* which can explain the higher resistance of its grains facing chemical and physical agents of scarification and of hot water (Patane and Gresta, 2006). In Mediterranean environments, grains of numerous species of leguminous must survive in hot and dry summers to allow seedlings, avoid temperatures potentially lethal and to delay germination until the next spring. Grains of those species developed a resistance mechanism to this stressful condition and complex machinery we can perceive when environmental conditions are favourable to break dormancy (Scippa et al., 2011). Collection, evaluation and valorisation of local populations of Medicago in Algeria began from 1971/1972 by Abdelguerfi. As a matter of fact, he identified 17 species belonging to different bioclimatic stages (Abdelguerfi, 1989). Although those species showed an interesting use in agriculture and a large adaptability with a wide range of environmental conditions (Chebouti and Abdelguerfi, 1999), little information is available in literature on characteristics of the grain's germination and techniques to break its seed coat imposing dormancy (Chabouti and Abdelguerfi, 1999). Several treatments had been proposed to reduce and improve tegument's hardness and rise germination rate in others species of leguminous notably Acacia (Danthu and Neffati, 2003). In this context this research aimed to evaluate response of grains' germination of Medicago originating of two different harvesting years (2004 and 2012) to different treatments of scarification, in order to promote their use in improvement of natural rangelands of steppes and Mediterranean areas.

MATERIALS AND METHODS

Vegetable material or seed come from collection of National Higher Agronomic School (NHAS). The code and weight of 1000 grains of used populations are shown in Table 1. This seed is harvested in 2004 and 2012. The storage of the seed of 2004 is made in an ordinary room requiring no conditioning. We wanted to test germination of this seed with one of year 2012. The studied species are classified according to the colour and size into three groups. Species with big grain (*Medicago intertexta, Medicago ciliaris*) of black colour, species with small grain (*Medicago polymorpha, Medicago truncatula)* of yellow-brown colour and species with middle size *Medicago muricoleptis* of black colour. The shape of all species is feeding nozzle.

Treatments of populations

(i) Seed treatment with sulphuric acid during 5 mn for the first test and during 180 mn for the second test. After we rinse under running water.
(ii) Passing of the seed through the drying oven at temperature of 115%c during 20 mn.
(iii) Isolation of an animal and introduction in its diet of 500 gr of pods by meal, twice a day during six days (*M. polymorpha, M. ciliaris* and *M. truncatula).* Then liberation of ovine and recuperation of faeces' seed by soaking in tap water and sifting.
(iv) Passing of the seed into boiled water (100°C) on the fire during 20 s.
(v) Passing of the seed in the freezer during 1 h, but for both steppe species during 5 days in the refrigerator.

It should be noted that seed of each population is put in small perforated closed cages which we have made with grid and sieve. Cages are denominated by codes of corresponding population. So

Table 2. Role of the seed coat on germination and survival of the species.

Populations and harvest year	Number of seeds	sprouts	%germination
Aus106/2004	8	0	0
S5 et S7/2004	12	0	0
Poly201/2004	8	7	87,5
Aus106/2012	4	0	0
S5 et S7/2012	8	0	0
Poly201/2012	8	8	100
I11/2012	20	1	5
Moyenne			27,5

during passing in chemical products, cages are gathered and put in a recipient containing the adequate product (acid, liquid nitrogen, boiled water or passing in freezer). After the timing, rinsing is made under tap running water. Drying is made in the open air. As for passing through the oven, seed is put in paper bags. Witnesses undergo no treatment.

Seedling after seeds treatment

After all treatments, seed (treated and witness) is sown in Petri boxes full of cotton. As for recuperated seed from faeces of ovine a second seedling is made, it is about of alveolus bearing a mixture of ground and compost to stimulate germination. It should be noted that seed without teguments is also sown to know their effect on the germination. Histological cuts have been performed on teguments by method of double coloration (methyl green and red congo). Observations have been made by optical microscope. The seed irrigation in Petri boxes is done with tap water. The rate calculation of the germination begins at the stopping of appearance of rootlets by counting number of germinated grains x 100 divided by the total number of grain contained in Petri box. Analysis of variance is carried out by statview software.

RESULTS AND DISCUSSION

As for grains without teguments, all studied species gave a germination rate of 0% except $Poly_{201}$ in the two study years which gave 100 and 87.5% of germination respectively for year 2012 and 2014 (Table 2). Also, the recuperated seed of ovine's faeces (Photos 1 and 2) gave 0% germination; we can say that passing of the seed in digestive tracks of ruminants has no scarifier effect. However; animal by its displacements may have a disseminator effect of the seed especially during its transhumance.

Analysis of the variance showed that storage years of seed have no influence on different rates of obtained germinations. Since, according to Tran and Kavavgh (1984) the waterproof coat protects embryo from unfavourable storage and environmental conditions and extend actively longevity of seeds. A combination of two mechanisms: closer shrinking of grains envelope cells and impregnation of cells with hydrophobic substances may exist and operating mechanisms may vary between different species. Observation of histological cuts by optical microscope, after passing of seeds in boiled water and after coloration in green of Methyl and in red Congo gave Photos 3 and 4. These photos showed three different layers responsible of protection and of dormancy of seeds. The best results are obtained by mechanical scarification (with sandpaper). The percentage of germination reaches until 100% (Table 3).

If we compare this method to other chemical and thermal ones, most of the results are lower than its Nonetheless, the results average of Figure 1 and Table 4 show that the passing in liquid nitrogen gives higher results close to results of scarification with sandpaper. While witness exceeds average of other methods, In fact, result of passing of the seed in sulphuric acid at 98% during 180 mn is approaching the witness. This result eventually may be improved by decreasing the pause time at 60 mn. Accroding to Patane and Gresta (2006) in *M. orbicularis* even after 2 mn at 100°C, 97.9% of grains remained more difficult to germinate. Exposure at ultra-low-temperature (-196°C) in liquid nitrogen finally improves germination and speed of grains germination of *M. Orbicularis* (Photos 5, 6 and 7). Chemical scarification with sulphuric acid was also efficient in the reduction of hard grains but has the higher concentration (70%) and the longest exposure time (60 mn) (Photos 8 and 9).

With regards to both steppe populations, their passing in refrigerator during five days gives a positive effect only for one but nothing for the other. That means 0% of germination for *M. truncatula* and 68% of germination for *M. polymorpha.* While witness scarified with sandpaper gave 69.84 and 90.57% respectively for *M. truncatula* and *M. polymorpha* (Photos 10 and 11).

Since the variance anyalisis showed no significant difference, thus does not influenceon result of different used tests; we take in consideration values of populations of both years as repetitions which gives $P<0.0001$ for all methods. Therefore, difference is very highly significant between species and between populations for the different tests (Table 5).

All seeds from a same lot do not present the same aptitude to the germination. This means that as seeds of a lot are dormant as germination is heterogeneous. Seeds considered as dormant are not necessarily seeds

Photo 1. Isolation of the animal and introduction pod 500 g per meal.

Photo 2. Recovery of semen by screening feces.

which do not germinate at all. Dormancies show up most often by a low and heterogeneous germination (Mullon and David, 1982). Causes of variation of the dormancy are various. First of all there are genetic, because dormancy varies according to species, within the same species depends on variety (Mullon and David, 1982).

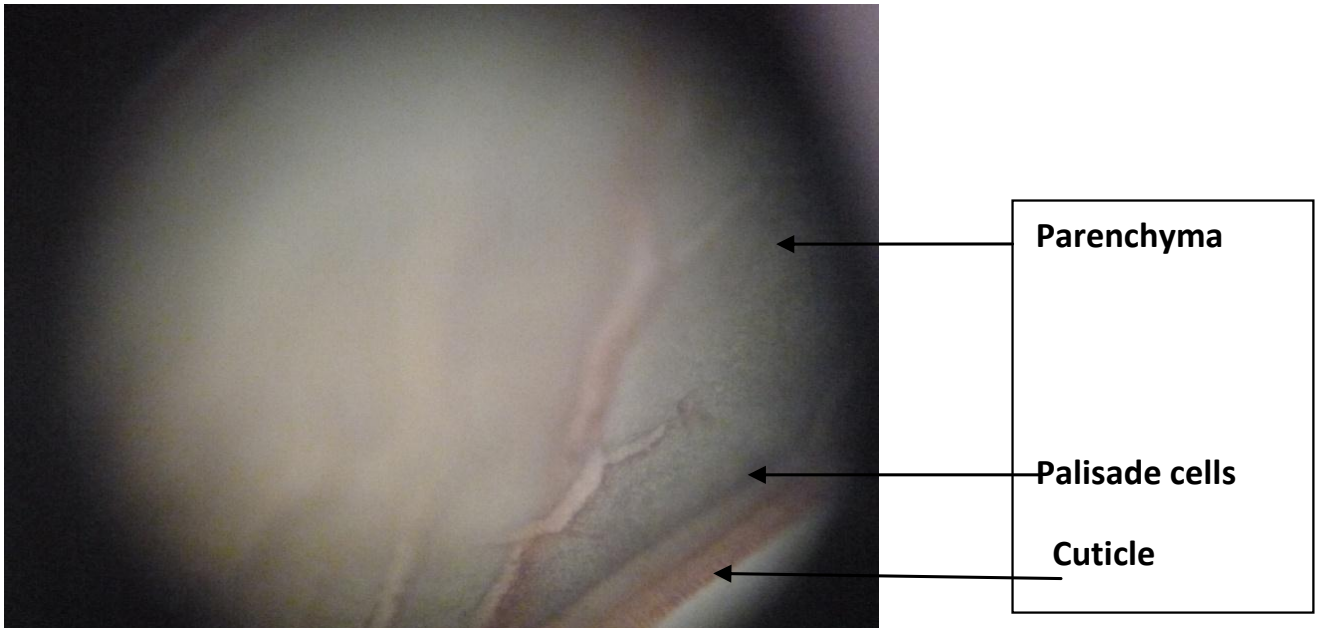

Photo 3. Histological cut at level of the seed tegument of *M. muricoleptis.*

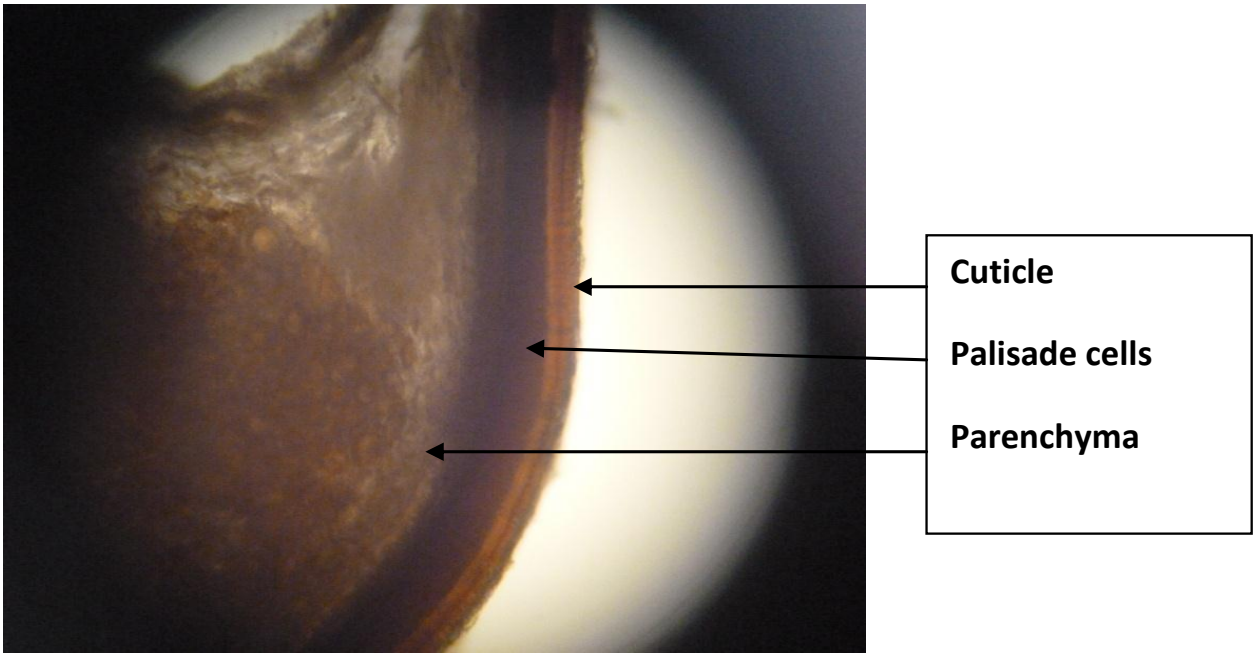

Photo 4. Histological cut at level of the seed tegument of *M. intertexta.*

The Medics do not germinate because their embryo has no possibility to be soaked. If we take the mechanical method, we noticed that this dormancy differs between species with big grain and small grain which that means: *M. ciliaris*, *M. intertexta* and *M. polymorpha*, *M. truncatula.* Within the small grains polymorph have a very high germination rate compared to *M. truncatula* (Table 2). However, within big grains, germination rate is very close between *M. ciliaris* and *M. intertexta.* Therefore, *Medicago* produces a strong proportion of dormant grains, also called “hard” grains which germinate after several years. For that reason it is not necessary to

Table 3. Results of two examples of scarification.

Harvest seed	Populations	scarification avec papier verre			scarification avec acide (5-6mn)		
		Number of seeds	Sprouts	% germination	Number of seeds	Sprouts	% germination
2004	Tr55	19	10	52,632	18	6	33,333
2004	Tr2012	14	13	92,857	21	2	9,524
2004	S5	9	9	100,000	42	0	0,000
2004	S7	15	14	93,333	38	0	0,000
2004	Poly205	15	14	93,333	24	6	25,000
2004	Poly60	32	32	100,000	9	0	0,000
2004	I756	24	24	100,000	45	3	6,667
2004	I52	24	24	100,000	28	2	7,143
2004	Aus106	25	25	100,000	55	2	3,636
2012	Tr55	18	18	100,000	8	2	25,000
2012	Tr201	63	45	71,429	3	2	66,667
2012	S5	11	11	100,000	20	0	0,000
2012	S7	16	12	75,000	20	5	25,000
2012	Poly205	26	23	88,462	18	1	5,556
2012	Poly60	17	17	100,000	13	3	23,077
2012	I756	16	16	100,000	23	6	26,087
2012	I52	16	16	100,000	16	1	6,250
2012	Aus106	16	14	87,500	14	1	7,143
Average of five species				91,919			15,005

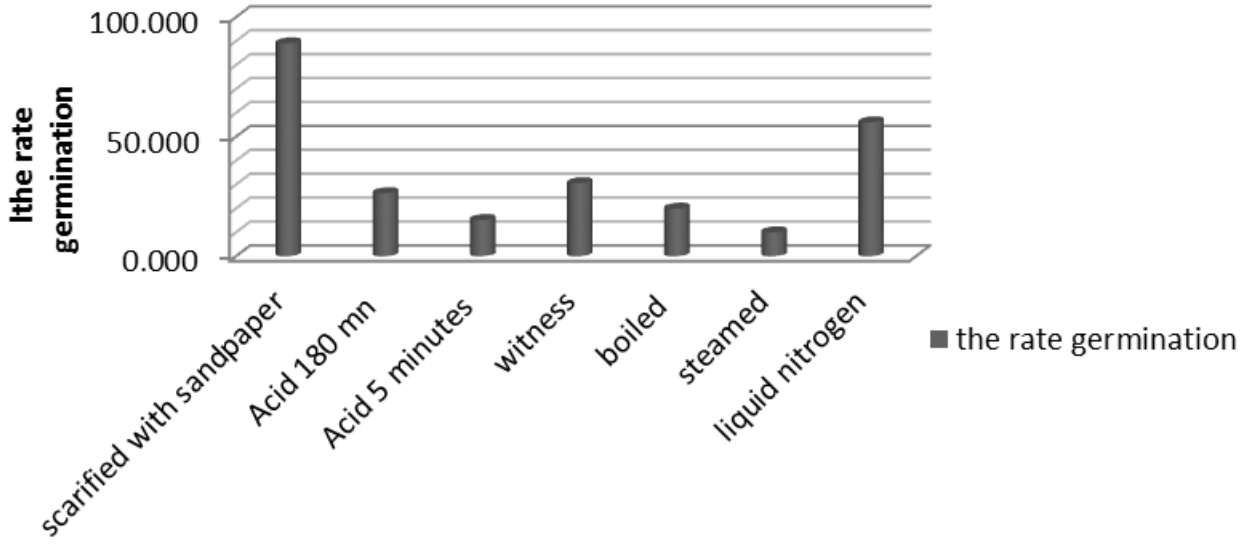

Figure 1. Germination rate of medics after different treatments of scarication.

reseed rangelands with medicago after implantations. Dormant grains ensure regeneration of rangelands during several years, in spite of seasonal rainfall fluctuations (FAO, 1997). A Medicago's rangelands characteristic consist in their production capacity of abundant supply of green forage richer in proteins during winter and spring with a duration varying according to species and cultivars of the same species. The question is to know if these species adapt better the African semi-arid surroundings. In our test on the field, we noted that crawling species with small grains such as *M. polymorpha* and *M. truncatula* sown in reduced space between grains, give interwoven plants in spider's web shape which can help contributing to erosive soils retention. We think they

Table 4. Germination results in % of Medics' populations after different tests of scarification (chemical, physical and mechanical).

Populations	Year	Witness	Scarified with sandpaper	Boiled	Steamed	5 mn Acid	180 mn Acid	Cold	Liquid nitrogen
Tr55	2004	25	52,632	-	3,571	33,333	11,765	0	89,66
T201	2004	36,111	92,857	4,167	0	9,524	0	25	78 ,45
S5	2004		100	11,111	8,333	0	16,129	2,941	20,97
S7	2004		90,476	14,286	5,263	2,632	0	9,524	23,9
Poly205	2004		93,333	11,429	1,754	25	8,824	33,333	63,83
PoLY60	2004		100	6,452	8,955	0	44,444	14,286	43,75
Aus106	2004		92,593	3,448	12,5	3,636	0	8,108	62,07
I756	2004		100	50	3,704	6,667	95,24	37,931	71,76
I52	2004		100	22,222	1,282	7,143	30,435	23,810	55,1
Tr55	2012		100	20,3704	19,44	25	27,273	28,571	68,42
T201	2012		71,429	12,5	23,256	66,67	28,125	24,138	85,2
S5	2012		100	10	6,122	0	-	13,636	29,41
S7	2012		75,000	14,286	23,529	25	59,091	9,524	32
Poly205	2012		46,939	6,897	3,226	5,556	80	20	70
PoLY60	2012		100	16,667	14,706	23,077	-	13,636	43,75
Aus106	2012		87,500	14,286	2,703	7,14	1,429	18,919	39,08
I756	2012		100	50	14,815	26,087	10,417	23,077	61,64
I52	2012		100,000	70	26,923	6,250	8,571	9,524	68,22
Average		30,556	89,042	19,889	10,005	15,151	26,359	17,553	56,002

should be sown in arid zones to improve soils structures through the contribution of organique mater vegetable and animal after pastures.

Environmental factors are known to influence the mother plant, provoking thus changes in characteristics of the progeny germination (grains). However, types of changes which may occur in grains, as they are developing under different environmental conditions, should also be taken in consideration. So, research is often necessary to *determine the relative importance* of each one of these two factors in dormancy variation of grains and germination of particular species (Baskin and Baskin, 1998).

Conclusion

The dormancies are very profitable to wild species, as they contribute ensuring their maintain in ecological conditions and under difficult climatic conditions. But sometimes there are important problems with cultivated species for which obtainment of germination necessitates research of adjusted treatments in each case. The dormancy is heterogeneous in Medicago, varies between species and within populations, the variance is high. Effect of physical treatment or chemical gives a germination rate which varies between 0 and 95.34% for I_{756} (acid 180 mn), 50% for $I_{756,\ 70}$% for I_{52} boiled and 66, 67% for Tr_{201} (acid 5 mn) while higher to 50% case of cold in steppes species. The same observation is made within populations. Thus, this is an inter-specific variation and inter-population.

Furthermore, both treatments which offer germinations rate potentially high and fast in all populations which are: Scarification with sand-paper and passing in liquid nitrogen. In spite of this latter gives lower result than the first one, it is more practical and easier to perform than all others means. It is about of a fast passing of a limited number of grains to be sown, but scarification with sand- paper, number of grains is

Photo 5. Bottle of liquid nitrogen.

Photo 6. Effect of liquid nitrogen on the cages containing the seed.

Photo 7. Seven days after sowing.

Photo 8. Seed in acid.

Photo 9. Rinçage seed.

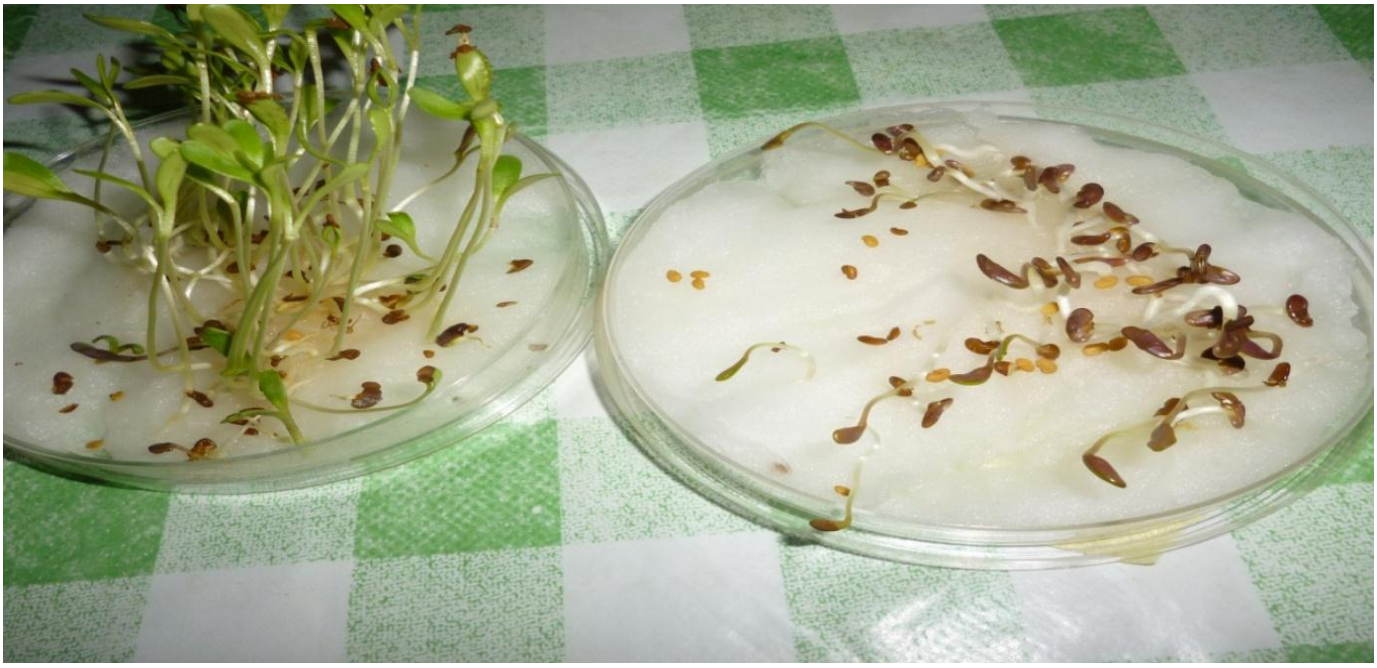

Photo 10. Germination of *M. polymorpha* right after refrigeration and left after scarification.

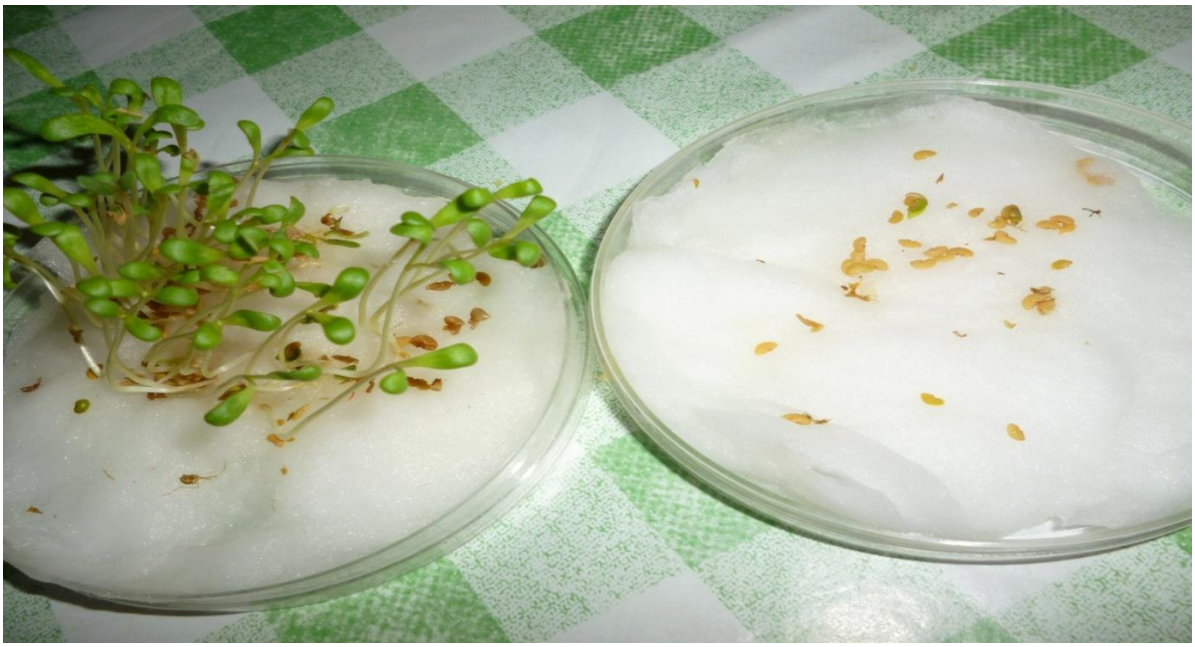

Photo 11. *M. truncatula* left after scarification and right after refrigeration has not germinated.

Table 5. Variance analysis of scarified Medics by different methods.

Parameter	Variance	DOF	P	Significance
Scarified seed	277,738	17	<0.0001	***
Boiled seed	350,159	16	<0.0001	***
Steamed seed	73,605	17	<0.0001	***
Passing in acid 5 mn	284,673	17	<0.0001	***
Passing in acid 180 mn	862,836	15	<0.0001	***
Passing in freezer 2 h	106,329	17	<0.0001	***
Passing in liquid nitrogen	440,899	17	<0.0001	***
Witness	61,727	1	<0.0001	***

DOF degree-of-freedom*** very highly significant.

limited and uses a long time. The germination percentage of these scarified populations by sand-paper in 2013 and sown in 2014 gave the following results: 21.92% of germination in ciliaris, 15% of germination in cultivar Gemalong, 18.5% in Truncatula, 26% in Intertexta and 7.5% in Granadensis. Therefore, we note that Intertexta keep their germination power more than others species.

Conflict of Interests

The authors have not declared any conflict of interests.

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Full Length Research Paper

Pesticide use in the production of Tomato (*Solanum lycopersicum* L.) in some areas of Northern Ghana

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Tomato (*Solanum lycopersicum* L.) is a very important vegetable used in almost all meals and is consumed in diverse ways. In Ghana, farmers and consumers of fruits and vegetables face immense risk of exposure owing to the use of toxic chemicals that are banned or restricted in the country or in other countries. The objective of this study was to ascertain farmers' access and use of recommended agro-chemicals for the production of tomatoes in three sampled farming communities in Northern Ghana. Sixty semi-structured questionnaires were administered and data analysed using Minitab Statistical package with T-test for significance. Various agro-chemicals were used by farmers which include: those not suitable for tomatoes production; unapproved or banned agro-chemicals and those suitable for tomatoes production. Communities which produced the "Burkina" variety used about 70% of the sampled pesticides compared with 30% for the "Wosowoso" variety. The agro-chemical most used (32.8%) was Dichlorodiphenyltrichloroethane (DDT) though banned from the Ghana registered list of pesticides. Farmers have access to, and use agro-chemicals for tomato production in the study areas. Farmers therefore need to be sensitized on the use of recommended and appropriate agro-chemicals and the hazards associated with the use for the crop, farmer and environment.

Key words: Banned, food safety, growth regulators, organochlorines, pesticides, tomato.

INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is a plant species from the Solanaceae family, which originated from the Americas. Like its close relatives, chili peppers and potato, tomato was probably introduced to Africa in the 16th century (Esquinas-Alcazar, 1981). Although definitive statistics on area and production of most crops are not readily available for many African countries, tomato is one of the most widely cultivated vegetable on the continent. The total production area in Africa increased from 159,593 ha in 1961 to 660,215 ha in 2007, and quantities produced increased from 1,968,812 tonnes in 1961 to 14,918,554 tons in 2007 (FAO, 2009). According to FAO (2009), the largest area and the highest production in Africa are found in Northern Africa,

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including Egypt, Morocco, and Algeria, and the smallest area and the lowest production in Southern and Central Africa, respectively (FAO, 2009). The average yields range from 6 t/ha in Central Africa to 34 t/ha in Southern Africa, with the southern region higher in productivity mainly because of South Africa (FAO, 2009). There has been an increase in area and production in individual countries over the past 50 years, but at different rates; productivity largely remained low. In Tanzania, the area increased from 1,400 ha in 1961 to 19,000 ha in 2007, but yield remained stagnant between 7.1 to 7.6 t/ha (FAO, 2009).

According to MoFA (2012), the total land size used for the production of tomato is 44.8 ha while 62.8 ha of land is used for the production of other vegetables in Ghana. Aside this large area of land used for the production of tomato, tonnes of tomato is imported from neighboring countries such as Burkina Faso and Togo to meet the demand for tomatoes. FAO (2015) estimated a produced quantity of 320,500 tonnes with a domestic quantity supply of 630,893 tonnes, an export of 4,828 tonnes and an import of 315,221 tonnes for tomato and tomato products in Ghana for the year 2011.

Tomato is a very important vegetable used in almost all meals either cooked or uncooked. The fruit may also be processed into juice, soup, ketchup, puree, paste or powder.

In Ghana, farmers and consumers of fruits and vegetables face immense risk of exposure owing to the use of toxic chemicals that are banned or restricted in the country or in other countries (Nasr et al., 2007). Wrong application techniques and time of spray of fruits and vegetables, badly maintained or totally unsuitable equipment and agrochemicals exacerbate these risks of residual deposition (Al-Wabel et al., 2011). Some agro-chemical residues in vegetables may however, be unavoidable even when sprayed in accordance with good agricultural practices (Uysal-Paha and Bilisli, 2006).

Research conducted for the past decade internationally and in Ghana points to the presence of pesticide residues in fruits and vegetables such as cabbage, onion, cucumber, lettuce, tomatoes, okra and pepper (Hanson et al., 2007; El-Nahhal, 2004; Hussain et al., 2002). Most of these pesticide residues are often classified as organochlorines or organophosphates with a preference for the organophosphorus for been less persistent, does not often bio accumulate, and are less hazardous to the farmer, consumer and environment.

Organochlorines are considered persistent organic pollutants (Pops), a category of chemicals that include nine organochlorides (aldrin, chloradane, DDT, dieldrins, endrin, heptachlor, hexachlorobene, mirex and toxphen) targeted by Stockholm convention in May 2001 which was aimed to eliminate their production and restrict or ban their use throughout the world (Lemairie et al., 2004). This can be explained in part by the long life of many organochlorines in the environment (dieldrins, DDT and its metabolites (DDD and DDE) can remain in the soil for decades) and long distance transport in wind and water current as well as food imports from countries that continue to use these pesticides. Inhalation and dermal contact are additional routes of exposure, both for individuals working directly with the pesticides and for children who are exposed to pharmaceuticals products, containing organochlorines such as head lice treatments. Infants are also exposed when organochlorine pesticides that have accumulated in their mother's bodies are passed to them in breast milk.

A higher proportion of pesticide poisonings and deaths occur in developing countries because there are inadequate protective clothing, and washing facilities; insufficient enforcement; poor labelling of pesticides; illiteracy; and insufficient knowledge of pesticides concentration in vegetables (Pimentel and Greiner, 1996). The objective of this study was to ascertain farmers' access and use of the recommended agro-chemicals for the production of tomatoes.

MATERIALS AND METHODS

Study area

Farmers were sampled from Pungu and Doba in the Kassena Nankana East District of the Upper East Region and Bunglung in the Savelugu/Nanton Municipality of Northern Region. These communities were sampled based on their dry season production of tomatoes which supplies the major cities such as Bolgatanga and Navrongo in the Upper East Region and Tamale in Northern Region when the supplies from the Southern part of Ghana are out of season. Farmers in Pungu and Doba produce the "Burkina" variety while farmers in Bunglung produce the variety "Wosowoso".

Data collection and analysis

Data was collected using semi-structured questionnaire, field visits, and interviews for farmers while one agro input shop per community was visited for the agro input market survey. Sample size of thirty farmers was used for each district and data analysed using T-test for significance in Minitab version 16. Secondary data on the pesticide register was accessed from the Environmental Protection Agency, Ghana in the identification of recommended, registered and banned pesticides.

RESULTS AND DISCUSSION

Pesticide used by farmers and availability

All sampled communities had a point of sale for agro-chemicals which were medium to small-scale. Various agro-chemicals and growth regulators were in use by farmers from the three sampled areas (Table 1). From Table 1, farmers used a combination of agro-chemicals suited for tomatoes, agro-chemicals not suited for tomatoes, unapproved and banned agro-chemicals affirming the assertion that consumers are exposed to

Table 1. List of agro-chemicals in use by farmers.

Chemical	%
***Dichlorodiphenyltrichloroethane (DDT)	32.8
Top Cop	18.8
Confidor	10.9
*Super force	9.4
*Harvest more	6.2
Kocide	4.7
*Grow force	3.1
Sulfa 80 WDG	3.1
Zap 2.5 EC	3.0
*Top harvest	1.6
Kombat	1.6
Lambda 25 EC	1.6
*Sidalco liquid	1.6
**Neem extract	1.6
Total	**100**

*Agro-chemicals not found on the EPA's register of pesticides; **Plant extract; ***Banned.

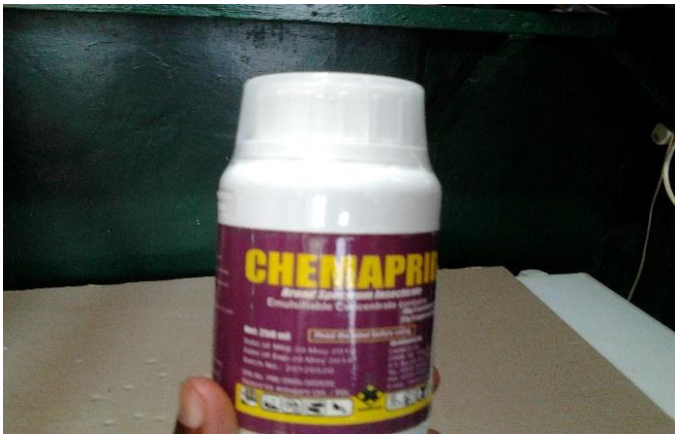

Figure 1. Chemaprid, a broad-spectrum insecticide sold to farmers as DDT.

pesticide residues through the consumption of tomatoes (Essumang et al., 2007). Communities which produced the "Burkina" variety used about 70% of the sampled pesticides compared with 30% for the "Wosowoso" variety.

The most used agro-chemical (32.8%) was Dichlorodiphenyltrichloroethane (DDT) which was classified as banned (EPA, 2013) and farmers did not believe DDT was banned. A follow-up visit to the agro-chemical shops further indicated that DDT was sold to some farmers in "second hand" containers unlabelled per the quantity of chemical required. It was also found that some agro-chemical outlets sell out broad-spectrum chemicals (Figure 1) to unsuspecting farmers as DDT, without considering the target crop.

This could be termed as causing economic fraud on users due to users' ignorance and inability to read the information on the label. About 50% of farmers disagreed DDT was banned for agricultural use. From a total of 13 agro-chemicals used in the study areas, five excluding the neem extract are approved and recommended (Zap 2.5 EC, Top Cop, Sulfa 80 WDG, Lambda 25 EC and Kombat) for tomato production. Confidor an insecticide for controlling capsid, bugs and insect pests in cocoa pods and Kocide 2000 a fungicide for the control of diseases in cocoa were found in use for tomato production in some sampled areas accounting for 10.9 and 4.7% respectively.

This could be considered as pesticide "misuse" even if it controlled the target disease or pest effectively as the two crops are physiologically varied and are processed and used differently. In addition, the Kocide was labelled as "packaged for Cocobod – Not for sale" (Figure 2), but found its way to the input sellers.

Growth regulators such as grow force, harvest more, sidalco liquid, super force and top harvest could not be found on the pesticide register but were used by farmers and were readily available at some agro-chemical shops. It was found that only pre-harvest pesticides were used in their production. It was also found that plant based control pesticides such as neem extract was not commonly used by farmers. Farmers argued that its preparation was tedious and time consuming.

Pesticide application practices

The stages for application of chemicals were not being followed by farmers. Chemicals were sometimes applied when diseases and pests/insects appear, and when produce are almost ready to be harvested. About 50% of the farmers often mix two or more agro-chemicals with the perception that it will ensure high efficacy. This practice can be attributed to the farmers' educational background as about 60% were not educated and did not know the health implications of using non recommended chemicals, application dose, time of application and the lethal phase before consumption. Surprisingly, 30% of farmers had separate plots for tomato production for their household use, where they do not apply agro-chemicals used for their commercial tomato farmlands. The farmers attributed the use of pesticides on their commercial tomato for public consumption based on the fact that consumers preferred good looking fruits at the point of sale. About 80% of farmers applied pesticides without protection (personnel, equipment, chemical and environment) and did not consider the time for most applications while 40% kept unused chemicals in bed rooms and kitchens for future use confirming with Al-Wabel et al. (2011). Most farmers derived and updated their knowledge and skills from friends, their continuous

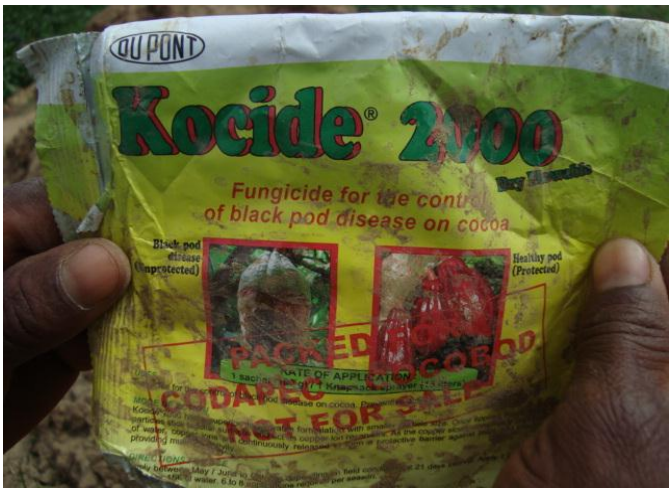

Figure 2. Used Kocide pouch left on the farm.

production and the use of the chemicals based on trial and error.

CONCLUSION AND FUTURE IMPLICATIONS

Farmers have access and are using agro-chemicals for tomato production in all three sampled communities with pesticides, weedicides and growth regulators been the most used. Most of these agro-chemicals were however, not meant for the cultivation of tomato nor vegetables and could result in the accumulation of pesticide residues. About 50% of the agro-chemicals in use were not found in the EPA's register of pesticides which sends a very worrying situation as to how these pesticides found their way onto the market. DDT, though banned could be found on the market in concealed "second hand" packaging materials and unlabelled while some farmers were been short-changed by agro-chemical dealers by selling out, general broad-spectrum agro-chemicals as DDT. Many farmers did not know that DDT was banned for some time now. Most farmers applied agro-chemicals without protection and kept pesticides in bed rooms and kitchens for future use.

To curb these problems associated the use of pesticides, input dealers, extension agents and farmers must be educated on the hazards related to the use of pesticides to themselves, the crops, consumers and the environment as a whole. Input dealers must engage qualified personnel who can provide technical and agronomic assistance to farmers in selecting the right type of chemical, applying them in right quantities and at the right time. Authorities with the responsibility to regulate the use and misuse of agro-chemicals must be proactive and check the influx of these banned chemicals on the market in order to save human lives, the environment and to prevent economic fraud on farmers.

The extension services division must be resourced to ensure extension agents recommend suitable agro-chemicals and also be able to reach out to farmers. The creation of plant clinics in communities will help farmers identify problems with crops and access professional solutions which would help reduce agro-chemical use and abuse in the farming communities in Ghana. A check list of approved and banned agro-chemicals should be supplied to input dealers and farmer groups.

Conflict of Interests

The authors have not declared any conflict of interests.

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Full Length Research Paper

Effect of weeds on yield loss of cassava plants in response to NPK fertilization

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The objective of this study was to assess yield loss of weed-infested cassava and the degree of interference of weeds on the crop productivity in response to the fertilization of NPK. The study was conducted at the Universidade Estadual do Sudoeste da Bahia, in the municipality of Vitória da Conquista, BA, with two different treatment groups that were evaluated in parcels with and without fertilizer. The treatments of the first group consisted of initial coexistence periods in which the cassava plants and weeds were put to live together: 35, 70, 105, 140 and 175 days after planting (DAP); in the second group, the cassava plants, initially, remain free from weeds during the same periods. Weeds were assessed every 35 days, from the 35 to 525 days after planting of cassava, determining the fresh mass of the identified species in the evaluated treatments with and without fertilizers. The characteristics of root yield, shoot weight, harvest index, dry mass of roots, starch content and flour production were evaluated 18 months after plantation. The predominant weeds in the experimental area were: *Panicum maximum*, *Brachiaria plantaginea*, *Sida rhombifolia*, *Pavonia cancellata*, *Portulaca oleracea*, *Cynodon dactylon* and *Setaria parviflora*. The interference of the weeds in the cassava yield was bigger when the crop was subjected to fertilization, in coexistence periods from the 35 days after planting; however, when cassava plants were kept in the absence or in coexistence with weeds up to 35 DAP, fertilization provided increase in crop productivity factors. The competition with weeds resulted in high losses in root yield of the cassava plant, thus, being necessary the control of the invading plants in the period between 35 and 175 DAP.

Key words: Coexistence period, competition, control, fertilizer, *Manihot esculenta*.

INTRODUCTION

Cassava plant (*Manihot esculenta* Crantz) grows under varied conditions of climate and soils, even the ones with low fertility (Carvalho et al., 2007b). Such characteristics enable the plant to be of expressive economic and social

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Table 1. Particle size and chemical analysis of the topsoil (0-20 cm) of the typical Dystrophic Yellow Latosol in the experimental area[1/]. Vitória da Conquista / BA, UESB (2015).

Particle size analysis (dag kg^{-1})			
Clay	**Silt**	**Coarse sand**	**Fine sand**	**Textural class**
29	1	56	14	Sandy Clay Loam

Chemical analysis										
pH	**P** [2/]	**K^+** [2/]	**H + Al** [3/]	**Al^{+3}** [4/]	**Ca^{+2}** [4/]	**Mg^{+2}** [4/]	**CTC_{total}**	**V**	**m**	**OM**
H_2O	**--- mg dm^{-3}---**		**---------- $cmol_c$ dm^{-3} ----------**					**------- % -------**		**dag kg^{-1}**
5.2	10.0	0.14	3.6	0.4	1.4	0.8	5.9	39	15	18.0

[1/]Results provided by Laboratory of Soil Analysis of the UESB; [2/] extractor Mehlich^{-1}; [3/] extractor Ca(OAC)2 0.5 mol L^{-1}, pH 7.0; [4/] extractor KCl 1 mol L^{-}.

importance in regions considered mostly unsuitable for farming, making the cassava plant an alternative of subsistence and generation of jobs and income in places where occur the least levels of human development index (HDI), in Brazil and in the world (Silva et al., 2014). Most of the cassava crops are concentrated in small farms, under rustic management, which is characterized as a low-input system, resulting in low yields of roots in Brazil (Alves et al., 2012), with low national average of 14.6 t ha^{-1} (IBGE, 2014), when it is compared to the productive potential of the crop, which can reach about 90 t ha^{-1} of tuberous roots (Cock et al., 1979). Cardoso et al. (2013) also add that among the main reasons for low productive index of the crop is the poor technology adoption in the farming system, low yielding varieties and, mainly, the competition with weeds.

Weed in cassava cultivation has been reported as one of the main factors affecting crop yield. According to Albuquerque et al. (2008), root yield can be reduced by more than 90% in absence of weed control. This is mainly due to a slow initial growth of cassava plants, which facilitates weed species development, favoring the competition for water, light, nutrients, carbon dioxide and physical space (Azevêdo et al., 2000). In addition, cassava harvest can occur up to two years after planting, when roots are delivered to processing industry (Silva et al., 2012). Because of long cultivation and the soil partial covering by the plant, several weed infestations can occur within the planting area, what might increase crop yield losses (Johanns and Contiero, 2006).

Although, the degree of interference of weeds in the crops depends on factors related to the crop such as, the weed community, the environment and the period in which they coexist (Silva et al., 2007). In this process, one of the factors more easily controllable, in practice, is the length of the coexistence period between crop and weeds (Pitelli, 1985). According to Carvalho (2000), the degree of this competition depends on the species, the population density and, mainly, the period in which they remain growing together. Therefore, identification of the most frequent species of weed is necessary because each one – according to the potential to establish in the area and the aggressiveness – can interfere differently with the cassava plant.

Among resources liable to weed and crop competition, nutrient extraction and accumulation appear to be a crucial feature when studying the entire weed community in competition with intermediate cycle crops, such as cassava (Albuquerque et al., 2012).

Fertilizers can be used to alter competitive relationships between crop and weed, favoring crop plants by changing weed community composition and density, since the species have different responses to nutrient inputs (Armstrong et al., 1993). Even though a large amount of knowledge and technological advances on crop mineral nutrition are available, there is a lack of that regarding infesting communities, what impairs the understanding of interfering factors in competition for nutrients between weed and crop plants (Procópio et al., 2005). Based on the above, this study aimed to identify the main species of weed found during cassava cultivation, and to evaluate its effect on crop productivity in response to NPK fertilization.

MATERIALS AND METHODS

The study was developed between January of 2013 and July of 2014, in the experimental area of the Universidade Estadual do Sudoeste da Bahia, *campus* Vitória da Conquista, BA. The geographic coordinates are 14°51' South and 40°50' West, 941 m of average altitude. The climate, in accordance with the Köppen classification is *Cwa* (Humid subtropical climate), with annual average rainfall of 741 mm. The soil of the experimental area was classified as typical Dystrophic Yellow Latosol (Oxisol) (EMBRAPA, 2006), of which main physicochemical characteristics are shown in the Table 1. Figure 1 depicts the climatic data that were obtained during the experiment, regarding the rainfall, relative humidity, maximum and minimum temperature.

Soil tilling consisted of plowing, harrowing and grooving. Fertilization of treatments was based on soil analysis and recommendation for cassava crop proposed by Nogueira and Gomes (1999). It was applied 40 kg ha^{-1} P_2O_5, directly into planting groove; and 70 kg ha^{-1} N and 30 kg ha^{-1} K_2O as top dressing, sixty

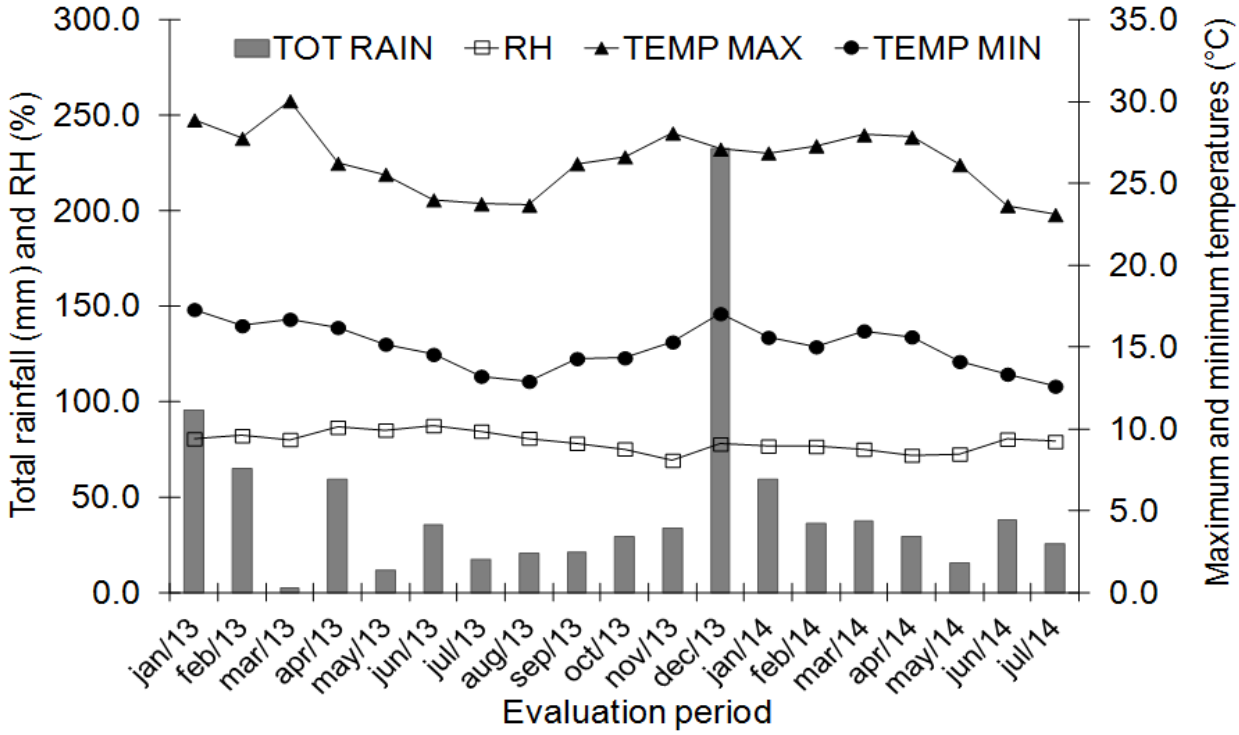

Figure 1. Monthly average rainfall (mm), relative humidity (%) and maximum and minimum temperature (°C), in the municipality of Vitória da Conquista-BA, in the period from January of 2013 to July of 2014.*Source: Instituto Nacional de Meteorologia – INMET/Vitória da Conquista, BA, 2014.

Table 2. Description of the periods of coexistence of weeds and cassava plants. Vitória da Conquista / BA, UESB (2015).

Treatment	Description
Control group 1[1/]	Crop always kept in areas free from weeds
CWD [3/] up to 35 DAP [4/]	Coexistence with weeds 35 days after planting
CWD up to 70 DAP	Coexistence with weeds 70 days after planting
CWD up to 105 DAP	Coexistence with weeds 105 days after planting
CWD up to 140 DAP	Coexistence with weeds 140 days after planting
CWD up to 175 DAP	Coexistence with weeds 175 days after planting
Control group 2 [2/]	Coexistence with weeds until the end of the cycle

[1]/ Crop free from weeds during the whole cycle; [2]/ crop cultivated with weeds during the whole cycle; [3]/ coexistence with weeds; [4]/ days after planting.

days after planting. In the second year, 60 kg ha^{-1} N and 60 kg ha^{-1} K_2O were applied as topdressing at the beginning of rainy season (December, 2013). For treatments without fertilizer application, it was considered soil natural fertility (Table 1).

The planting was manually conducted in January, 2013, where variety of Caitité was taken, which is a bitter variety, with approximately from 2 to 3 cm of diameter, 20 cm of length and seven buds. The spacing was 1.0 m between rows and 0.6 m between plants; each plot consisted of four lines of 8.4 m of length and 4.0 m of width, totaling 33.6 m^2. The usable area of the plot was represented by two central lines, leaving out 0.6 m in each extremity with frontal borders, with a usable area of 14.4 m^2.

The experiment consisted of two treatment groups, which both were evaluated in plots with and without fertilizers, and four repetitions. In the first group, it was assessed with the different periods of coexistence of weeds and cassava plants, as specified in Table 2. After each period of coexistence, the crop remained free from competition with weeds due to manual weeding.

In the second group, the treatments consisted of different periods of weed control that were performed by manual weeding, maintaining the crop free from interference of weeds after the planting, as shown in Table 3.

Table 3. Description of the periods of control of weeds in cassava crop field. Vitória da Conquista / BA, UESB (2015).

Treatment	Description
Control group 1[1/]	Crop always kept in areas free from weeds
FWD [3/] up to 35 DAP [4/]	Free from weeds 35 days after planting
FWD up to 70 DAP	Free from weeds 70 days after planting
FWD up to 105 DAP	Free from weeds 105 days after planting
FWD up to 140 DAP	Free from weeds 140 days after planting
FWD up to 175 DAP	Free from weeds 175 days after planting
Control group 2 [2/]	Crop always kept in areas with weeds

[1]/ and [2]/ Crop free from and with weeds during the whole cycle, respectively; [3]/ free from weeds; [4]/ days after planting.

The assessments of weeds were performed on the 35, 70, 105, 140, 175, 210, 280, 315, 350, 385, 420, 455, 490 and 525 days after the planting (DAP) of cassava. During these evaluations, weeds were collected through sampling. For that, a metallic square measuring 0.5 m x 0.5 m (0.25 m^2) was randomly thrown on the usable area of the plot with and without fertilization. Weeds situated within the sampled areas were cut at ground level and, afterwards, they were taken to the laboratory, where the identification, counting and weighing of the fresh mass of the species were conducted.

During the harvest, manually performed 18 months (June, 2014) after the planting, were evaluated the following characteristics: root yield (t ha^{-1}), shoot weight (t ha^{-1}), harvest index, dry mass of the roots (%), starch content (%) and flour production (%). In order to measure the root yield, roots were collected in the usable area; then, they were cleaned and weighed on a precision scale at 0.1 g. The shoot (leaves and stem) was separated by sectioning at a height of approximately 15 cm from the soil; after, the green mass was quantified. The harvest index was evaluated by using the formula: HI = root weight ÷ (root weight + weight of the shoot). The characteristics of dry mass of the roots and starch content were determined by the hydrostatic weighing (Grossmann and Freitas, 1950), considering zero for values below three kilograms of root. The root yield was determined through the method proposed by Fukuda and Caldas (1987).

A randomized block design with four replications was used in this experiment, with the treatments of each plot arranged in a 7 x 2 factorial scheme, in which there were seven initial periods of coexistence or weed control, evaluated with and without fertilization. The obtained data related to the assessment of the crop was subjected to variance analysis, by the F-test at 5% of probability and the means were compared to each other through the Tukey's test at 5% of probability.

Regarding the data from the assessment of the weed population, the treatments were distributed in a 15 x 2 factorial scheme (fifteen periods of evaluation and two fertilization conditions), using the F-test to compare the quadratic means and, and the means that had been compared by the Tukey's test at 5% of probability. In order to quantify the results, a regression analysis was performed, selecting the significant models ($F \leq 0.05$), with the coefficient of determination being above 60%. The statistical analysis of the data was carried out by using the software ASSISTAT, beta version 7.7.

RESULTS AND DISCUSSION

By surveying the weed population, 50 species were identified, which were classified in 39 genera and 15 botanic families. The main families, in regard to the number of species, were: Malvaceae (fourteen), Asteraceae (eight), Poaceae (seven) and Fabaceae (five), which represent 68% of the species found (Table 4). Similar results were found in surveys conducted by Otsubo et al. (2002), Albuquerque et al. (2008) and Guglieri et al. (2009), which also emphasized the mentioned families as being those of utmost number of species of weeds in cassava fields.

The composition of the weed community was considered to be heterogeneous in comparison to the survey conducted by Albuquerque at al. (2014), who by evaluating the occurrence of weed in cassava fields located in the cerrado (tropical savanna ecoregion of Brazil) of Roraima (Boa Vista, RR), reported a population of 27 weed species, distributed in 21 genera and 8 families. Conversely, in the phytosociological survey conducted by Huziwara et al. (2009), in the municipality of Campos de Goytacazes/RJ, it was identified 10 species that belong to 9 genera and 9 families of invasive plants of cassava fields.

Regarding the fresh mass produced by the infesting community, it was observed an increase of 47.6% in the treatments with fertilizers (12,390.51 gm^{-2}) in comparison with the treatments without fertilizers (8,393.28 g m^{-2}) (Table 4), indicating that the application of fertilizers to cassava crops favors the growth of weeds, which may lead to larger losses in the production of roots due to the competition. According to Cruz and Pelacani (1993), among the effects caused by the presence of weeds, the shade provided by species that grew more rapidly in the initial growth phase of the crop appears to be more relevant, because as the percentage of shading on cassava grows, the plant height increases, without increasing the accumulation of biomass in the stem and with reduction of the leaf area index. These authors conclude that, with less light exposure, the dry matter of the stem and leaves and the root yield are impaired. As a consequence, shading promotes delayed growth and decrease in the growth rate of the tuberous root.

Table 4. Species of weed that occur in cassava fields, organized by family, scientific name, Brazilian common name and fresh mass in the treatments with and without fertilization. Vitória da Conquista / BA, UESB (2015).

Family/Species	Brazilian common name	Fresh mass (gm^{-2})		
		F [1/]	NF [2/]	Total
Amaranthaceae				
Amaranthus retroflexus	Caruru-gigante	16.59^{A}*	1.96^{B}	18.55
Chenopodium carinatum	Anserina-rendada	29.39^{B}	100.66^{A}	130.05
Asteraceae				
Acanthospermum australe	Carrapicho-rasteiro	253.41^{A}	212.37^{A}	465.78
Acanthospermum hispidum	Carrapicho-de-carneiro	2.56^{A}	0.47^{B}	3.03
Bidens pilosa	Picão-preto	5.88^{B}	11.86^{A}	17.74
Blainvillea rhomboidea	Picão-grande	114.44^{A}	147.65^{A}	262.09
Emilia fosbergii	Falsa-serralha	71.09^{A}	28.38^{B}	99.47
Eupatorium ballotifolium	Picão-roxo	229.71^{A}	266.98^{A}	496.69
Siegesbeckia orientalis	Botão-de-ouro	0.39^{A}	0.62^{A}	1.01
Synedrellopsis grisebachii	Agrião-do-pasto	3.66	—	3.66
Boraginaceae				
Heliotropium indicum	Crista-de-galo	1.55^{A}	2.34^{A}	3.89
Brassicaceae				
Lepidium virginicum	Mentrusto	2.35^{A}	0.25^{B}	2.6
Commelinaceae				
Commelina benghalensis	Trapoeraba	269.55^{A}	1.11^{B}	270.66
Euphorbiaceae				
Chamaesyce hyssopifolia	Burra-leiteira	3.01^{A}	0.78^{B}	3.79
Euphorbia prostrata	Quebra-pedra-rasteira	1.93	—	1.93
Fabaceae				
Aeschynomene denticulata	Angiquinho	2.12^{A}	1.22^{B}	3.34
Crotalaria incana	Chocalho-de-cascavel	—	12.32	12.32
Senna obtusifolia	Fedegoso	61.51^{A}	59.03^{A}	120.54
Stylosanthes viscosa	Vassourinha	—	0.24	0.24
Zornia reticulata	Alfafa-do-campo	—	9.41	9.41
Malvaceae				
Gaya pilosa	Guanxuma	—	29.48	29.48
Herissantia crispa	Mela-bode	—	1.6	1.6
Herissantia tiubae	Malva-de-bode	0.46	—	0.46
Malvastrum coromandelianum	Falsa-guanxuma	58.94^{A}	30.03^{B}	88.97
Pavonia cancellata	Malva-rasteira	469.74^{A}	520.53^{A}	990.27
Pavonia sidifolia	Vassoura	42.83^{B}	109.93^{A}	152.76
Sida carpinifolia	Malva-baixa	38.25^{A}	6.24^{B}	44.49
Sida cordifolia	Malva-branca	317.63^{A}	217.99^{A}	535.62
Sida glaziovii	Guanxuma-branca	2.59	—	2.59
Sida rhombifolia	Guanxuma (vassourinha)	951.06^{A}	1016.27^{A}	1967.33
Sida santaremnensis	Guanxuma	1.38	—	1.38
Sida spinosa	Guanxuma-de-espinho	96.47^{A}	109.96^{A}	206.43
Sida urens	Guanxuma-dourada	24.24^{A}	8.2^{B}	32.44
Waltheria indica	Malva-branca	0.2^{B}	1.73^{A}	1.93

Table 4. Contd.

Molluginaceae				
Mollugo verticillata	Molugo	6.79^{A}	4.59^{A}	11.38
Nyctaginaceae				
Boerhavia diffusa	Agarra-pinto	28.24^{A}	9.41^{B}	37.65
Passifloraceae				
Passiflora cincinnata	Maracujá-do-mato	4.44	—	4.44
Poaceae				
Brachiaria plantaginea	Capim-marmelada	2527.21^{A}	1617.08^{B}	4144.29
Cenchrus echinatus	Capim-carrapicho	315.21^{A}	239.16^{A}	554.37
Cynodon dactylon	Grama-seda	497.54^{A}	252.67^{B}	750.21
Digitaria horizontalis	Capim-colchão	10.31^{B}	30.19^{A}	40.5
Panicum maximum	Capim-colonião	4452.05^{A}	2409.8^{B}	6861.85
Rhynchelytrum repens	Capim-favorito	169.27^{A}	68.58^{B}	237.85
Setaria parviflora	Capim-rabo-de-raposa	262.47^{B}	408.49^{A}	670.96
Portulacaceae				
Portulaca oleracea	Beldroega	594.47^{A}	253.01^{B}	847.48
Portulaca mucronata	Onze-horas	—	2,37	2.37
Rubiaceae				
Diodia teres	Mata-pasto	383.38^{A}	72.19^{B}	455.57
Richardia scabra	Poaia-do-cerrado	44.94^{A}	55.17^{A}	100.11
Solanaceae				
Solanum americanum	Maria-pretinha	28.2^{A}	12.96^{B}	41.16
Solanum erianthum	Caiçara	0.29^{B}	52.89^{A}	53.18
	Total	**12390.51**	**8393.28**	**20783.14**

*Means followed by the same letter in the row do not differ significantly from one another by the Tukey's test at 5% of probability. [1]/ and [2]/ Cultivation with and without fertilization, respectively.

The largest production of fresh mass of weeds was found from the 350 days after the planting of cassava, which is the beginning of the local rainy season (Figure 1); in these periods, the fertilized crop stood out from the rest by registering the maximum production between the 385 and the 455 days after the planting, increasing on average about 43% of fresh mass of the infesting community in relation to the unfertilized crop (Figure 2). Therefore, cassava fertilizations increased fresh mass production of weed, notably during the second cropping year; time when nutrient competition might not be harmful to cassava plants, since they already have well-formed shoot and root.

Despite being less expensive, weed control during second year can be difficult, because crop shoot has already been formed, which makes it difficult to enter into the field (Peressin and Carvalho, 2002). In this case, between the two cycles, the crop is in physiological rest. Falling leaves and plant reduced metabolic activity characterize this phase and its duration is related to environmental conditions especially. Therefore, it is during this period that a new infestation starts, which was also observed in this study, mainly in fertilized treatments from 350 DAP. It is therefore necessary to control these plants, to avoid possible losses and to facilitate crop harvesting (Silva et al., 2012).

The species with higher production of fresh mass were: *Panicum maximum* (33.02% in relation to the total mass), *Brachiaria plantaginea* (19.94%), *Sida rhombifolia* (9.47%), *Pavonia cancellata* (4.76%), *Portulaca oleracea* (4.08%), *Cynodon dactylon* (3.61%) and *Setaria parviflora* (3.23%) (Table 4). In regard to the percentage of total fresh mass produced in the treatments with and without fertilization, these species represented 78.72 and 77.17%, respectively (Figure 3). Concerning the responses of these species to fertilization, *P. maximum*, *B. plantaginea* and *C. dactylon* (Poaceae) had significant increases in fresh mass when fertilizer was applied, representing 84.74; 56.28 and 96.91%, respectively. Contrarily, *S. parviflora* had higher

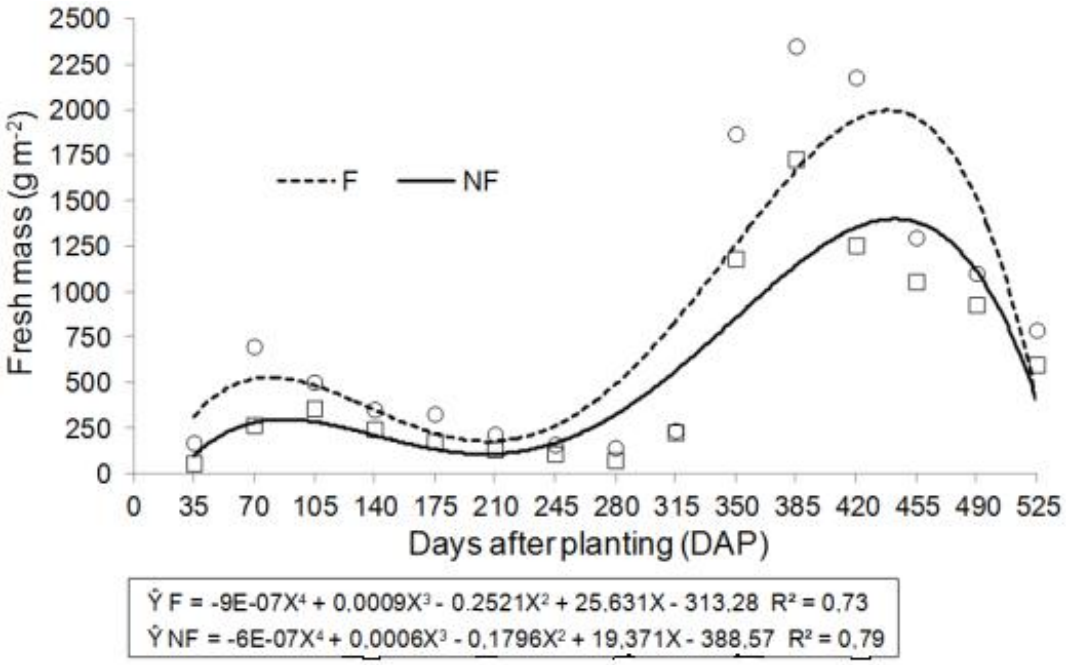

Figure 2. Fresh mass (g m^{-2}) of the shoot of weeds in the cassava crop between the 35 and the 525 days after the planting (DAP), in the assessed treatments with (F) and without (NF) fertilization. Vitória da Conquista / BA, UESB (2015).

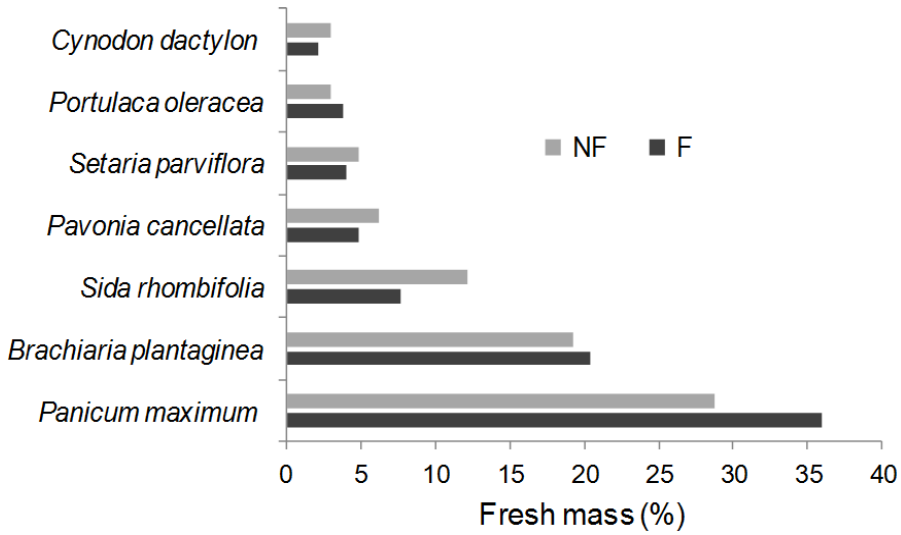

Figure 3. Average production of fresh mass (% in relation to the total mass) of species of weeds that prevail in the cultivation of cassava with (F) and without (NF) fertilization. Vitória da Conquista / BA, UESB (2015).

production of fresh mass in the non-fertilized treatments, reducing this rate in 35.74% for fertilized ones. In the family of Malvaceae, *S. rhombifolia* and *P. cancellata* were unresponsive to fertilization, showing a sensitive reduction in the fresh mass in the fertilized cultivation (6.42 and 9.76%, respectively), without differing, however, from the unfertilized cultivation. As for *P. oleracea* (Portulacaceae) the fertilization provided an increase of 135% in yield of fresh mass in comparison with the unfertilized cultivation (Table 4). Such results demonstrate that weed responses to fertilizers are variable with regards to fresh mass production.

According to Brighenti and Oliveira (2011), some weed species have greater efficiency to use fertilizers to grow faster, increasing the competition against crop. Within the weeds of greater occurrence in the experimental area, the species *P*. *maximum*, *B*. *plantaginea* and *S*. *rhombifolia* stood out, of which exhibited higher percentage of fresh mass than the remaining, representing 64 and 60% of the total fresh mass measured in the treatments with and without fertilization, respectively (Figure 3).

The species *P*. *maximum*, commonly known as "capim-colonião", exhibited high values of fresh mass in the shoot, being responsible for 35.93% of the fresh mass in the fertilized cultivation and 28.71% in the unfertilized one (Figure 3); suggesting high power of competitiveness due to its high biomass producing capacity in comparison to the other species. Its occurrence was recorded in some periods of the first cycle of crop, however, with reduced production of fresh mass. Nevertheless, from the 350 days after planting the cassava, phase characterized by the beginning of the local rainy season (Figure 1), it was observed a significant increase in the production of fresh mass in the plant shoot.

Among the probable factors that are responsible for the occurrence and elevated production of fresh mass in "capim-colonião", the following stand out: the presence of propagules of the species in seedbanks in the area, defoliation of cassava plants, fertilizer supply and beginning of the rainy season; these were the conditions that, certainly, favor the establishment and the development of *P*. *maximum* in the area, since the species is very light-demanding, fertility and soil moisture.

B. plantaginea, commonly known as "capim-marmelada", exhibited a good adaptation and aggressiveness in the cultivated area, being found in all the assessments of the infesting community; representing 20.39% of the total fresh mass obtained in the fertilized cultivation and 19.26% in the unfertilized cultivation (Figure 3). This Poaceae, originated from Africa, has the seed as the main way of propagation, which is characterized by exhibiting primary dormancy in the maturation process (Lorenzi, 2008); hence, the germination is distributed throughout the time, which impairs its control (Kissmann, 1997).

The high competing potential of the "capim-marmelada" was also verified in the study performed by Aspiazú et al. (2010), in which by evaluating the efficiency in watering cassava plants in coexistence with weed, it was verified that the *B. plantaginea* is very efficient at using water, principally for having a metabolism C_4, and it remains competing with the cassava even under temporary conditions of shortage of water.

The Poaceae family is highlighted as one of the most important group in cassava fields. Pinotti et al. (2010) identified the species *Brachiaria decumbens* and *Digitaria horizontalis* are economically important in cassava fields in the municipality of Pompéia-SP. Whereas Albuquerque et al. (2014), in a phytosociological study on the cassava crop in the cerrado (tropical savanna ecoregion of Brazil) of Roraima, concluded that the species that exhibited the highest values of dry mass were the Poaceae *Digitaria sanguinalis*, *Brachiaria brizantha*, *B. decumbens* and *Brachiaria humidicola*. As reported by Maciel et al. (2010), many species of the Poaceae family are perennial and produce a large quantity of seeds, increasing their dissemination and colonization of different environments.

The species *S*. *rhombifolia,* commonly known as "guanxuma", exhibited widespread occurrence in the experimental area, being recorded in every assessment of the infesting community; accounting for 7.67% of the total fresh mass that was produced by the fertilized cultivation and 12.1% in the unfertilized one (Figure 3). Such occurrence may be due to its high potential of infestation since this species exhibits high yield of seeds and easy dispersion.

The yield of fresh mass of "guanxuma" was larger in the second cycle of the crop, after the 350 days after planting of cassava, however, without considerable differences among the obtained values in the cultivation with and without fertilizers; demonstrating that the occurrence and production of fresh mass of this species are independent of the supply of fertilizer to the crop. Conversely, this performance can be explained by the higher efficiency of the Poaceae *P*. *maximum* and *B. plantaginea* in using the fertilizer supplied to the crop and the environment, and, consequently, in the production of fresh mass; which may have not favored the growth of the *S*. *rhombifolia* in the fertilized cultivation, in relation to the unfertilized one.

S. rhombifolia occurs in annual and perennial crops, being highly competitive due to its radicular system, which can reach 50 cm in depth (Kissmann and Groth, 2000; Lorenzi, 2008). Reports indicated that this plant can yield up to 28.2 thousand seeds per m^{-2} in only a cycle in the summer as a weed in soybean fields (Fleck et al., 2003). Within the species of "guanxuma" that occur in Brazil, it is considered the most disseminated species and its control is the most problematic, regardless the agricultural environment (Constantin et al., 2007). It was also reported as weed in cassava fields by Azevêdo et al. (2000) and Albuquerque et al. (2008), corn fields (Macedo et al., 2003), sugarcane fields (Oliveira and Freitas, 2008) and soybean field (Voll et al., 2005).

Generally, the application of fertilizers on cassava fields, performed as to support the crop that is suffering detrimental effects of weeds, also favored the production of fresh mass of weeds, chiefly, in the second year of the crop cycle; therefore, depending on the intensity of the

Table 5. Average yield (Y) and shoot weight (SW) of the cassava, variety Caitité, in different periods of coexistence with weeds. Vitória da Conquista / BA, UESB (2015).

Treatments	Y (t ha^{-1})	SW (t ha^{-1})
Control group 1 [1/]	26.40a*	8.79a
CWD [3/] up to 35 DAP [4/]	23.70^{a}	7.35^{a}
CWD up to 70 DAP	12.84^{b}	3.40^{b}
CWD up to 105 DAP	6.21^{bc}	2.32^{bc}
CWD up to 140 DAP	2.68^{c}	0.99^{bc}
CWD up to 175 DAP	0.87^{c}	0.37^{c}
Control group 2 [2/]	0.11^{c}	0.06^{c}
CV (%)	25.71	29.07

*Means followed by the same letter in the column do not differ significantly from one another by the Tukey's test at 5% of probability. [1]/ and [2]/ Crop free from and with weeds during the whole cycle, respectively; [3]/ coexistence with weeds; [4]/ days after planting.

competition in this phase, the crop might be negatively affected, because according to Procópio et al. (2005), depending on the management, the application of macronutrients may benefit more the species of weed than the crop.

Regarding the first group of treatments (periods of coexistence of crop and weeds), the root yield and the shoot weight of the cassava plant were not influenced by the fertilizer provided to the crop, being only influenced by the different periods of coexistence with the weed community (Table 5). By evaluating the yield, it was observed the decline in root production when the periods of coexistence of weeds and crop were similar or superior to 70 days between the planting and the weeding. The yield loss was about 90% when the first weeding was executed after the 140 days of coexistence of crop and weeds (Table 5).

The highest productivities of roots were observed in the control group free from competition and in the treatment in which the coexistence of crop and the weeds occurred up to 35 days after the planting (Table 5). Similar results to the ones found in this study were also verified by Carvalho et al. (2004), as they found out that the cassava crop, in the municipality of Cruz das Almas, Bahia, can coexist with weeds for a period from 20 to 30 days after the sprouting, about 35 to 45 days after planting, without significant loss in the production of roots.

The weeding that was performed after 70 days of coexistence of crop and weeds resulted in losses of 51% in productivity of cassava, in relation to that obtained in the cultivation kept free from weeds during the whole cycle (Table 5). Alcântara et al. (1983) also reported the that weeding on the 60 days after the sprouting showed reduction in the root and shoot yield, which accorded with the data obtained by Carvalho et al., (1990).

According to Johanns and Contiero (2006), in a study carried out in Marechal Cândido Rondon-PR, it was found out that the competition of weeds and cassava, cultivar "Fécula Branca", between the 60 and 90 days after the planting reduces significantly the yield of the crop, concluding that, in this period, there is more competition for resources. Albuquerque et al. (2008), in Viçosa-MG, reported that the cassava crop, cultivar "Cacauzinha", is more sensitive to competition in periods of coexistence between the 25 and 75 days after the planting. Biffe et al. (2010), in Maringá-PR, found out that weeding performed up to 100 days after planting considerably increased the production of cassava roots, cultivar "Fécula Branca".

The results verified in this study in which the lowest productivities were obtained in the treatments with weeding done after 140 days of coexistence and in the control group without weeding (Table 5), accord to some authors, which the cases where is reported the greater interference of weeds, the productivities are below 10% of the results obtained in the weeded control (Carvalho et al., 1993; Moura, 2000; Johanns and Contiero, 2006; Silva et al., 2012).

In shoot weight, it was noted that periods of coexistence with weeds from the 70 days after planting cassava considerably reduced the production of the plant shoot, and, consequently, the plant growth (Table 5). This small growth of the cassava shoot contributes to the decrease in photosynthetic tissue and, eventually a larger accumulation of carbohydrates in the roots, reducing the final yield of the crop (Viana et al., 2001).

In both cultivation, with and without fertilizers, the harvest index of the crop exhibited significant reduction from the 105 days of coexistence with weeds, with the less expressive results found in the treatments with CWD up to 175 DAP and in the control group with no weeding; however, despite the losses due to competition, superior indexes were observed in the unfertilized cultivation, more specifically in the treatments CWD up to 70 DAP, up to 105 DAP and in the control group kept in the among the weeds during the whole cassava cycle, in relation to the fertilized

Table 6. Means of harvest index (HI) and dry mass of cassava roots (DMR), variety Caitité, in different periods of coexistence with weeds, which were evaluated in fertilized and unfertilized cultivations. Vitória da Conquista / BA, UESB (2015).

Treatments	HI		DMR (%)	
	F [5]	NF [6]	F	NF
Control group 1[1]	0.75^{aA}*	0.75^{aA}	33.55^{aA}	31.88^{aB}
CWD [3] up to 35 DAP [4]	0.74^{aA}	0.78^{aA}	33.81^{aA}	32.2^{aB}
CWD up to 70 DAP	0.73^{aB}	0.80^{aA}	31.65^{bA}	32.85^{aA}
CWD up to 105 DAP	0.53^{bB}	0.67^{bA}	21.86^{cB}	33.04^{aA}
CWD up to 140 DAP	0.52^{bA}	0.48^{cA}	21.54^{cA}	21.63^{bA}
CWD up to 175 DAP	0.24^{cB}	0.25^{dB}	0.0^{dB}	10.44^{cA}
Control group 2 [2]	0.0^{dB}	0.22^{dA}	0.0^{dA}	0.0^{dA}
CV (%)	6.27		3.55	

*Means followed by the same letter (lowercase in the columns and uppercase in the rows) do not differ significantly from one another by the Tukey's test at 5% of probability. [1]/ and [2]/ Crop free from and with weeds during the whole cycle, respectively; [3]/ coexistence with weeds; [4]/ days after planting; [5]/ and [6]/ cultivation with and without fertilization, respectively.

cultivation (Table 6).

In relation to the percentage of dry mass of roots, in the fertilized cultivation, the losses were recorded starting from the 70 days of coexistence of crop and weeds; whereas in the unfertilized cultivation, these losses were retarded, being only verified in periods of coexistence starting from the 140 days after the planting cassava. In both cases, the least impressive results were verified in the treatments CWD up to 175 DAP and in the control group without weeding.

In the fertilized cultivation, it was recorded that the percentages of dry mass were higher than the ones found in the unfertilized cultivation, in the control group free from weeds and in the initial period of coexistence of crop and weeds (CWD up to 35 DAP). In the unfertilized cultivation, relating to the fertilization, higher percentages were obtained in longer periods of coexistence, namely the treatment CWD up to 105 DAP and up to 175 DAP (Table 6).

These results suggest that the fertilization with NPK alters the degree of competitiveness between the crop and the weed community, favoring the growth of species of weed after the 35 days after the planting of cassava, leading to the retardation of the beginning of the formation of roots and, consequently, the accumulation of dry mass. A similar behavior was observed by Pereira et al. (2012), in which was found out that while evaluating the growth of cassava and weeds in response to phosphorous fertilization the fertilization tends to contribute to a faster growing rate for weeds than for cassava. To Carvalho et al. (2007a), there is a greater development of the roots in the initial periods of the development cycle of weeds, resulting in an increase of nutrient uptake by radicular interception, leading to a quick accumulation of nutrients by the roots and, consequently, a lower availability for the crop.

In respect to the starch content and the flour production, as similar as the results observed for the dry mass of roots, in the fertilized cultivation, it was verified a decrease of these values when the periods of coexistence of crop and weeds were the same or superior to 70 days between the planting and the weeding. In cultivation without fertilization, the losses were recorded only after the 140 days of coexistence of crop and weeds. In both cultivation, the lowest results of starch content and flour production were observed in the treatments CWD up to 175 DAP and in the control group without weeding (Table 7). The results are in accordance with Silva et al. (2012), which affirmed that the presence of weeds in competition with the cassava crop may reduce the percentages of dry mass and starch content in the roots.

The effect of fertilization in the control group free from weeds and in the treatment CWD up to 35 DAP were recorded higher percentages of starch content and flour production, than the unfertilized cultivation. Conversely, in the treatments of coexistence of crop and weeds (up to 105 DAP and up to 175 DAP), in the unfertilized cultivation, it was obtained results that were higher than those found in the fertilized cultivation (Table 7); indicating that the effects of competition are sharp when the cultivation of cassava is submitted to fertilization.

Regarding the root yield that was obtained in the second group of treatments (initial periods of weed control), it was verified that there was no significant interaction between the periods of weed control and the fertilization, occurring only isolated effects of these factors (Figure 4).

By evaluating the productivity of cassava in the different periods of weed control, it was verified that the cultivation free from weeds during the whole cycle has the greater results, with 26.4 t ha^{-1}, 37% more than the productivity found in the treatment free from weeds up to the 175 days after the planting of cassava; showing, in this case, that the management of the crop in clean areas during the whole cycle leads to the maximum

Table 7. Means of starch content (SC) and flour production (FP) of cassava roots, variety Caitité, in different periods coexistence with weeds, assessed in fertilized and unfertilized cultivation. Vitória da Conquista / BA, UESB (2015).

Treatments	SC (%)		FP (%)	
	F [5]	NF [6]	F	NF
Control group 1 [1]	28.90^{aA}*	27.23^{aB}	26.33^{aA}	24.10^{aB}
CWD [3] up to 35 DAP [4]	29.16^{aA}	27.55^{aB}	26.68^{aA}	24.52^{aB}
CWD up to 70 DAP	27.00^{bA}	28.20^{aA}	23.79^{bA}	25.40^{aA}
CWD up to 105 DAP	18.76^{cB}	28.39^{aA}	16.87^{cB}	25.65^{aA}
CWD up to 140 DAP	18.44^{cA}	18.53^{bA}	16.44^{cA}	16.57^{bA}
CWD up to 175 DAP	0.0^{dB}	8.89^{cA}	0.0^{dB}	7.78^{cA}
Control group 2 [2]	0.0^{dA}	0.0^{dA}	0.0^{dA}	0.0^{dA}
CV (%)	4.14		6.16	

*Means followed by the same letter (lowercase in the columns and uppercase in the rows) do not differ significantly from one another by the Tukey's test at 5% of probability. [1]/ and [2]/ Crop free from and with weeds during the whole cycle, respectively; [3]/ coexistence with weeds; [4]/ days after planting; [5]/ and [6]/ cultivation with and without fertilization, respectively.

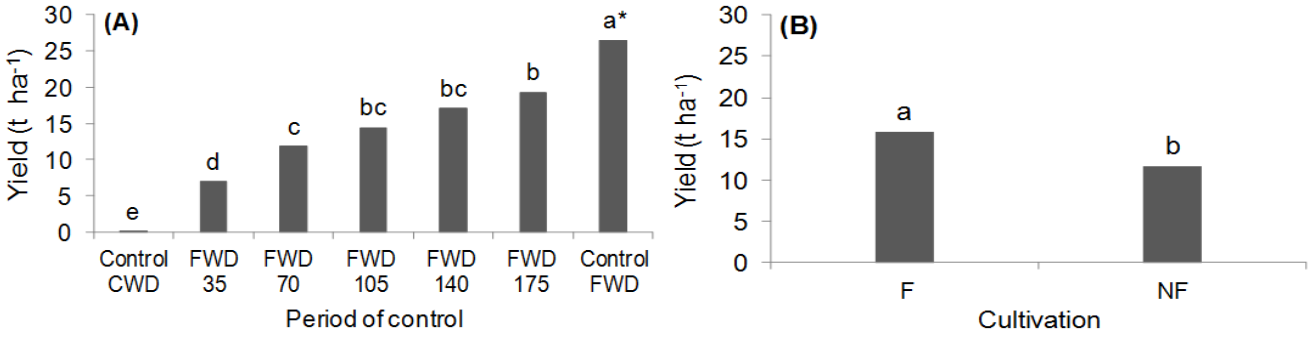

Figure 4. Means of yield of cassava, variety Caitité, in different periods of weed control (A), evaluated in cultivation with (F) and without (NF) fertilization (B). Vitória da Conquista / BA, UESB (2015).*Columns followed by the same letter do not differ from one another by the Tukey's test at 5% of probability. Control group CWD and control group FWD – Cultivation with and without weeds during the whole cycle, respectively; FWD – Cultivation free from weeds from the 35 to 175 days after planting of cassava.

economical potential of yielding roots (Figure 4A). The differences in productivity found within the treatments are probably due to a recurring infestation of weeds in the second year of the crop cycle (350 DAP) as a result of the beginning of the local rainy season (Figure 1), resulting in restarting the growth of weeds, as observed in the increase in fresh mass in this period (Figure 2), and the resumption of the competition with the crop. Therefore, in cassava fields with cycles longer than a year, it must be planned strategies of weed control that also cover the second year of the crop cycle.

Different results were found by Carvalho et al. (2004), in which, by studying the cultivar "Cigana preta" (BMG 116), in the municipality of Cruz das Almas, Bahia, indicate the necessity of weed control up to 135 days after planting the cassava. However, the periods of weed control are not absolute and they vary a lot, as it depends on factors that affect the production system, such as the cultivation spacing, species and population density of weeds, used cultivar, farm management, and hydric regime, among others. Therefore, the periods of weed control must be analyzed considering the location, environment characteristics and the management of the cassava field (Silva et al., 2012).

The yield of the cultivation that was kept with weeds during the whole cycle was severely affected, with losses exceeding 95% in comparison with the one free from weeds (Figure 4A). These losses are in accordance with the ones reported by Peressin et al. (1998) and Mattos and Cardoso (2005), as they affirmed that the losses in root yield as a result of competition with weeds during the cultivation of cassava may reach more than 90%, depending on the length of the coexistence and population density of weeds.

The fertilization boosted root yield by 35% in comparison with the unfertilized cultivation (Figure 4B), indicating that the improvement in soil fertility directly affects the cassava productivity. A similar effect was observed by Cardoso et al. (2005), which by studying the effect of nitrogen fertilization (0, 50, 100, 200, 300

Table 8. Means of weight of shoot (WS) and harvest index (HI) of cassava, variety Caitité, in different periods of weed control, evaluated in fertilized and unfertilized cultivation. Vitória da Conquista / BA, UESB (2015).

Treatments	WS (t ha^{-1})		HI	
	F [5/]	NF [6/]	F	NF
Control group 1 [1/]	9.44^{aA}*	8.14^{aA}	0.75^{aA}	0.75^{aA}
FWD [3/] up to 35 DAP [4/]	3.56^{bcA}	2.90^{bcA}	0.64^{abA}	0.53^{abA}
FWD up to 70 DAP	4.50^{bA}	3.53^{bcA}	0.70^{aA}	0.73^{aA}
FWD up to 105 DAP	5.65^{abA}	4.21^{abcA}	0.76^{aA}	0.73^{aA}
FWD up to 140 DAP	6.12^{abA}	5.07^{abA}	0.76^{aA}	0.75^{aA}
FWD up to 175 DAP	7.17^{abA}	6.14^{abA}	0.75^{aA}	0.75^{aA}
Control group 2 [2/]	0.0^{cA}	0.11^{cA}	0.0^{bA}	0.22^{bA}
CV (%)	35.29		22.81	

*Means followed by the same letter (lowercase in the columns and uppercase in the rows) do not differ significantly from one another by the Tukey's test at 5% of probability. [1]/ and [2]/ Crop free from and with weeds during the whole cycle, respectively; [3]/ free from weeds; [4]/ days after planting; [5]/ and [6]/ cultivation with and without fertilization, respectively.

and 400 kg ha^{-1} of N) on the agronomic characteristics of cassava, variety "Sergipe", in the municipality of Vitória da Conquista-BA, it was verified that the increase in root yield in relation to the amount of nitrogen applied, reaching up to 22.1 t ha^{-1} when it is applied 400 kg of N, which represents an increase of 20% in comparison with the unfertilized cultivation.

Likewise, Alves et al. (2012), by evaluating the effect of NPK (0, 200, 400 and 600 kg ha^{-1}) on the root yield, in the municipality of Moju-PA, it was concluded that in sandy soils and of low fertility, the variety of cassava "Paulozinho" responded linearly to the growing amounts of mineral fertilizer NPK. Nonetheless, Fidalski (1999) verified that only the phosphate fertilization (0, 30, 60, 90 and 120 kg ha^{-1} of P_2O_5) contributed to the increase in root yield of cassava, cultivar "Fibra", when it is submitted to NPK fertilization in sandy soils in the northwest of Paraná.

Relating to the weight of the shoot and the harvest index of the crop, it was observed that there were no differences between the fertilized and the unfertilized cultivation for the periods of weed control, demonstrating that the application of fertilizers to the crop has no influence on these variables (Table 8).

In both cultivations, with and without fertilization, it was verified that as the periods of weed control increased, there was an increase in weight of the shoot of cassava, with the higher result observed in the treatment that was kept free from weeds during the whole crop cycle, although not differing from the treatments FWD up to 105 DAP, up to 140 DAP and up to 175 DAP (Table 8). The data accord with the ones obtained by Albuquerque et al. (2008), in which the cultivation kept free from weeds exhibited the highest weight of the shoot, though not differing from the treatments free from weeds up to 75, 100 and 125 days after planting the cassava, cultivar "Cacauzinha". According to Silva et al. (2012), the production of the plant shoot is of great importance for the cassava production, supplying material to the propagation and to the production of forage, besides of being the part of the plant responsible for absorbing light and providing photoassimilates to the roots.

As for the harvest index, in the fertilized and unfertilized cultivation, except for the treatment kept with the weeds during the whole crop cycle, the remaining treatments did not differ from one to another (Table 8). In accordance with Conceição (1986), a good harvest index should be at least of 60%; therefore, the treatments exhibited a good harvest index. The fact that the harvest index remained unchanged for the majority of the treatments shows that the reduction of the plant shoot is closely linked to the decrease in root yield, that is, the weeds influenced the general growth of the cassava crop.

According to Moreira et al. (2014), the harvest index, alone, does not provide the precise information about the performance of the cassava plant, as the high values of this index can either be obtained by the increase of the production of roots or by the decrease of production of the shoot.

For the variables of dry mass of roots, starch content and flour production regarding the second group of treatments, there was no influence of fertilization, only of the different periods of weed control (Table 9). On the contrary, Cardoso et al. (2005) studied the effects of nitrogen fertilization on these characteristics, however, there were minor increases, which according to the authors, it is somewhat not economical advantageous.

When the crop was kept without weeding during the whole cycle, the production of roots remained below 3.0 kg ha^{-1}, making it inviable to determine the percentages of dry mass, starch content and flour production; while the remaining treatments did not differ from one to another (Table 9). These results showed that these characteristics are not influenced by the presence of weeds up to the 175 days after planting cassava. Similar

Table 9. Means of dry mass of roots (DMR), starch content (SC) and flour production (FP) of cassava roots, variety Caitité, in different periods of weed control. Vitória da Conquista / BA, UESB (2015).

Treatments	DMR (%)	SC (%)	FP (%)
Control group 1 [1/]	32.71[a]*	28.06[a]	25.21[a]
FWD [3/] up to 35 DAP [4/]	30.39[a]	28.01[a]	25.14[a]
FWD up to 70 DAP	32.66[a]	25.74[a]	22.11[a]
FWD up to 105 DAP	30.82[a]	26.17[a]	22.69[a]
FWD up to 140 DAP	32.32[a]	27.67[a]	24.68[a]
FWD up to 175 DAP	31.87[a]	27.22[a]	24.08[a]
Control group 2 [2/]	0.00[b]	0.00[b]	0.00[b]
CV (%)	7.13	8.35	12.60

*Means followed by the same letter in the column do not differ significantly from one another by the Tukey's test at 5% of probability. [1]/ and [2]/ Crop free from and with weeds during the whole cycle, respectively; [3]/ free from weeds; [4]/ days after planting.

results were obtained by Albuquerque et al. (2008), which the periods of weed control (25, 50, 75, 100 and 125 DAP) did not differ from each other for these characteristics when cultivating the cassava cultivar "Cacauzinha".

According to Correia et al. (2005), cassavas have about 30 to 40% of dry mass in tuberous roots however, the amount of this content relies on factors, such as: variety, age of plants, soil, planting conditions, climate and plant health. The content of dry mass is the characteristic that determines the higher or lower industrial yield of roots, since it is directly related to several products that are derived from cassava (Vidigal-Filho et al., 2000).

Generally, by analyzing the experiment as a whole, it was verified that although cassavas kept free from weeds during the whole cycle have exhibited the higher productivity, it is known that this is not a recommended practice, as maintaining the soil totally free from weeds, there is an increase in erosion and also a higher production cost. According Aguiar to et al. (2011), a significant parcel of the production cost can be attributed to the weed control, which can vary according to the species of weed and their population density. Therefore, this study, for the studied conditions, the first weeding should be performed near to 35 DAP and the last ones, near to 175 DAP.

The fertilization of cassava, when competing with weeds, provided an increase of productive factors (dry mass, starch content and flour production) when the period of coexistence was up to 35 DAP, though, in longer periods of coexistence after the planting, the losses by competitions were greater than the losses found in the unfertilized cultivation.

Conclusions

In conclusion it can be said that weed community found in cassava plantation was heterogeneous with 50 species distributed into 39 genera and 15 botanic families, and predominated by *P. maximum*, *B. plantaginea*, *S. rhombifolia*, *P. cancellata*, *P. oleracea*, *C. dactylon,* and *S. parviflora*.

The use of fertilizers in cassavas provided more production of fresh mass of weeds, mainly, in the second year of the crop i.e., from the 350 DAP, which was found maximum in *P. maximum* and *B. plantaginea.*

The interference of weeds on the cassava yield was higher when the crop was subjected to fertilization with NPK, in periods of coexistence starting from the 35 DAP. Moreover, the fertilization promoted the increase of factors related to productivity like, dry mass, starch content and flour production either in absence or in coexistence with weeds up to 35 DAP. The competition with weeds resulted in high losses of root yield of cassava; thus, it is necessary to control these plants in a period situated somewhere between the 35 and 175 DAP.

Conflict of Interests

The authors have not declared any conflict of interests.

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Morphological characterization of pearl millet hybrids [*Pennisetum glaucum* (L.) R. Br.] and their parents

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The present study was undertaken to characterize pearl millet [*Pennisetum glaucum* (L.) R. Br.] hybrids and their parents on the basis of morphological descriptors with the objective to identify key diagnostic characters of the genotypes. A set of 24 pearl millet genotypes (7 hybrids and their 17 parental lines) was evaluated in randomized block design (RBD) with three replications at two locations, Chaudhary Charan Singh Haryana Agricultural University (CCS HAU) Hisar and Regional Research Station (RRS) Bawal (Rewari) under irrigated and rainfed conditions, respectively, during kharif, 2011. Observations were recorded for 28 morphological and yield characters. All genotypes were classified into different groups based on each character. Nodal pubescence, nodal pigmentation, spike shape, spike density, spike tip sterility, sheath pubescence and spikelet glum colour distinguished all the 24 genotypes by assigning them key diagnostic features that would certainly help the plant breeders, seed growers and seed certification agencies to use these diagnostic characters. Hybrids HHB 216, HHB 226, HHB 117 could be differentiated by bristle length, spikelet glume colour and spike tip sterility. Majority of the morphological characters found to be dominant in the hybrids were contributed by the male parent.

Key words: Pearl millet, morphological descriptor, characterization, key diagnostic character.

INTRODUCTION

Pearl millet [*Pennisetum glaucum* (L.) R. Br.] is extensively cultivated for grain as well as fodder in the dry areas of north western and southern India. It is the fourth most important staple food crop after rice, wheat and sorghum primarily grown for grain and fodder production (Yadav et al., 2007). Pearl millet has an ability to grow in environments of low and erratic rainfall, high temperature and low soil fertility. Therefore, pearl millet is the main source of food and fodder for the poor farming communities which are habitant to these environments. With its ability to adopt to diverse agro ecological conditions, it occupies a unique position in the world agriculture.

Pearl millet has wide genetic diversity which is of little value unless it is characterized, evaluated and documented properly to enhance its utilization in crop improvement. In order to utilize effectively the available genetic diversity, the material must be properly

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Table 1. List of pearl millet hybrids and their parents studied.

S/N	Hybrids	Year of release	Female parent	Male parent	Maintainer
1	HHB 94	2000	ICMA 89111	G 73-107	ICMB 89111
2	HHB 117	2004	HMS 7A	H 77/ 29-2	HMS 7B
3	HHB 67 Imp	2005	ICMA 843-22	H 77/833-2-202	ICMB 843-22
4	HHB 197	2007	ICMA 97111	HBL 11	ICMB 97111
5	HHB 216	2010	HMS 37A	HTP 3/13	HMS 37B
6	HHB 223	2010	ICMA 94555	HBL 11	ICMB 94555
7	HHB 226	2010	ICMA 843-22	HBL 11	ICMB 843-22

characterized and catalogued. With the proliferation of newly developed varieties in important cultivated crops, the task of establishing the identity of these varieties and maintaining their seed lots has become a major concern. Characterization of varieties is thus of significance for the purpose of establishment and verification of identity and assessment of varietal purity for seed production and certification. Apart from this, characterization of pearl millet genotypes is also required for their protection under Plant Varieties Protection (PVP) legislation, because varietal testing for distinctness, uniformity and stability (DUS) is the basis for grant of protection of new plant varieties under the Protection of Plant Varieties and Farmers' Rights Act, 2001 (PPV and FR Act, 2001). Thus, the objective of present study was to characterize pearl millet hybrids and their parents using morphological descriptors developed as DUS descriptors (All India Coordinated Pearl Millet Improvement Project (AICPMIP), 2006).

MATERIALS AND METHODS

The present investigation was carried out at the research farm of The Department of Genetics and Plant Breeding, Chaudhary Charan Singh Haryana Agricultural University, Hisar under irrigated condition and Regional Research Station, CCS Haryana Agricultural University, Bawal (Rewari) under rainfed conditions. The experiment was conducted during the kharif season, 2011. The seed material for the present study consisted of 24 genotypes (7 hybrids, 6 CMS lines, 6 B lines, 5 restorers) of pearl millet as given in Table 1. A set of 24 genotypes was evaluated in randomized block design (RBD) with three replications under irrigated conditions. The plot size was one row of 4 m length with row to row distance of 50 cm and plant to plant 15 cm. Recommended agronomic practices were followed to raise the crop. The observations were recorded on 5 plants in each replication at specified stages of crop growth period when the characters under study had full expression. Pooled data from both locations were used to classify the genotypes. Flow charts were also prepared depicting key diagnostic characters related to genotypes.

RESULTS AND DISCUSSION

The identification of a pearl millet genotype is established by using a set of morphological characteristics. These characteristics are also useful to establish distinctness, uniformity and stability of the new variety, based on which the variety is given protection. In the present study, 28 morphological characteristics listed in the DUS test guidelines and morphological descriptors for pearl millet were explored for varietal description of pearl millet. Among the 24 pearl millet genotypes, considerable variation was observed between the genotypes for all the important attributes under study at both locations except anthocynin coloration of first leaf sheath, number of nodal tillers, spike girth, stem thickness, leaf mid rib colour and ligule hair. All the qualitative traits, as expected, were found similar over the locations (Kumar et al., 1993). The characterization of pearl millet hybrids and their parents under study is presented in Table 2. Key diagnostic characters of each genotype are assigned through flow chart in Figure 1.

In the present study, sheath pubescence was absent in all the genotype except HMS 7A and HMS 7B. On the basis of number of productive tillers, genotypes were classified in three categories: low, medium and high, having four, sixteen and four genotypes, respectively. On the basis of anther colour, genotypes were classified in two groups: yellow and brown, all the genotypes had yellow anther colour except HHB 94, ICMB 97111 and G 73-107 which possessed brown anther colour. One hybrid (HHB 67 Improved) and eight parental lines showed absence of nodal pubescence whereas all other fifteen genotypes showed presence of nodal pubescence. Among 24 genotypes of pearl millet under study, eleven expressed green nodal pigmentation, four expressed brown nodal pigmentation, six had red nodal pigmentation, two had purple nodal pigmentation and G 73-107 exhibited whitish nodal pigmentation. All the genotypes showed green internode pigmentation except ICMA 97111 and ICMB 97111 which exhibited purple internode pigmentation. All the genotypes showed complete ear head exertion type except HMS 37A and HMS 37B (partial ear head exertion). Only HHB 117 had brown spikelet glum colour whereas all the other genotypes of pearl millet under study were having green spikelet glum colour. HHB 67 improved and its male parent H77/833-2-202 showed purple bristle colour whereas HHB 197, HHB 223, HHB 226, HHB 216 and

Table 2. Characterization of pearl millet hybrids and their parents on the basis of morphological descriptors.

Plant descriptors	Expressions	No. of genotypes	Genotypes Hybrids	Parental lines
Anthocyanin coloration of first leaf sheath	Absent	24	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 94, HHB 117, HHB 216	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
Productive tillers	Low (2-3)	4	-	ICMA 94555, ICMB 94555, HMS 37A, HMS 37B
	Medium (3-6)	16	HHB 67 Improved, HHB 197, HHB 223	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
	High (>6)	4	HHB 226, HHB 94, HHB 117, HHB 216	-
Nodal tillers	No tillers	24	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 94, HHB 117, HHB 216	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
Sheath pubescence	Present	2	-	HMS 7A, HMS 7B
	Absent	22	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 94, HHB 117, HHB 216	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
Leaf length (cm)	Medium(51-60)	3	-	H 77/833-2-202, H 77/29-2, G 73-107
	Long (61-70)	13	HHB 226	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, HTP 3/13
	Very long (>70)	8	HHB 67 Improved, HHB 197, HHB 223, HHB 94, HHB 117, HHB 216	ICMA 94555, ICMB 94555
Leaf width (cm)	Narrow (<3)	3	-	H 77/833-2-202, G 73-107, HBL 11
	Medium (3-4)	15	HHB 67 Improved, HHB 197, HHB 223, HHB 94, HHB 226	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, HMS 37A, HMS 37B, HMS 7A, HMS 7B
	Broad (>4)	6	HHB 117, HHB 216	ICMA 94555, ICMB 94555,HTP 3/13, H 77/29-2
Anther colour	Yellow	21	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 117, HHB 216	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, HTP 3/13
	Brown	3	HHB 94	ICMB 97111, G 73-107
Number of nodes	Low (< 11)	22	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 94	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
	Medium (11-15)	2	HHB 117, HHB 216	-

Table 2. Contd.

Nodal pubescence	Present	15	HHB 197, HHB 223, HHB 226, HHB 94, HHB 117, HHB 216	ICMA 843-22, ICMB 843-22, HMS 37A, HMS 37B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
	Absent	9	HHB 67 Improved	ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 7A, HMS 7B
Internode pigmentation	Green	22	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 94, HHB 117, HHB 216	ICMA 843-22, ICMB 843-22, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
	Purple	2		ICMA 97111, ICMB 97111
	Whitish	1	-	G 73-107
Nodal pigmentation	Green	11	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 117, HHB 216	ICMA 94555, ICMB 94555, HBL 11, H 77/29-2, HTP 3/13
	Brown	4	HHB 94	ICMA 843-22, ICMB 843-22, H 77/833-2-202
	Red	6	-	HMS 37A, HMS 37B, HMS 7A, HMS 7B, ICMA 89111, ICMB 89111
	Purple	2		ICMA 97111, ICMB 97111,
Ear head exertion	Complete	22	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 94, HHB 117, HHB 216	ICMA 843-22, ICMB 843-22, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13, ICMA 97111, ICMB 97111
	Partial	2	-	HMS 37A, HMS 37B
Spike length (cm)	Small (11-20)	15	-	ICMA 843-22, ICMB 843-22, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13, HMS 37A, HMS 37B
	Intermediate (21-30)	9	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 94, HHB 117, HHB 216	ICMA 97111, ICMB 97111,
Spikelet glume colour	Green	23	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 94, HHB 216	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
	Brown	1	HHB 117	-
Bristle length (cm)	Short (< 0.5)	2	HHB 67 Improved	H 77/833-2-202
	Medium (0.5-2.0)	6	HHB 197, HHB 223, HHB 226, HHB 216	HBL 11, HTP 3/13
Spike girth (mm)	Medium (16-30)	24	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 94, HHB 117, HHB 216	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
Bristle colour	Purple	2	HHB 67 Improved	H 77/833-2-202
	Brown	6	HHB 197, HHB 223, HHB 226, HHB 216	HBL 11, HTP 3/13

Table 2. Contd.

Spike shape	Conical	13	HHB 223, HHB 94	ICMA 843-22, ICMB 843-22, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, G 73-107
	Cylindrical	5	HHB 67 Improved, HHB 197	ICMA 97111, ICMB 97111, HBL 11
	Candle	5	HHB 226, HHB 117, HHB 216	H 77/29-2, HTP 3/13
	Lanceolate	1	-	H 77/833-2-202
Spike tip sterility	Present	16	HHB 197, HHB 223, HHB 94, HHB 216	ICMA 843-22, ICMB 843-22, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, HTP 3/13
	Absent	8	HHB 67 Improved, HHB 226, HHB 117	ICMA 97111, ICMB 97111, H 77/833-2-202, H 77/29-2, G 73-107,
Spike density	Compact	9	HHB 67 Improved, HHB 197, HHB 226, HHB 117, HHB 216	ICMA 843-22, ICMB 843-22, HMS 7A, HMS 7B,
	Semi-compact	4	HHB 223, HHB 94	H 77/29-2, HBL 11
	Loose	11	-	ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, H 77/833-2-202, G 73-107, HTP 3/13
Plant height (cm)	Short (101-150)	6	-	ICMA 94555, ICMB 94555, HMS 7A, HMS 7B, ICMA 843-22, ICMB 843-22,
	Medium (151-200)	10	HHB 67 Improved,	ICMA 89111, ICMB 89111, HMS 37A, HMS 37B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
	Tall (201-250)	5	HHB 197, HHB 223, HHB 226,	ICMA 97111, ICMB 97111
	Very tall (>250)	3	HHB 94, HHB 117, HHB 216	-
Stem thickness (cm)	Medium (0.6-1.0)	24	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 94, HHB 117, HHB 216	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
Leaf mid rib colour	White	24	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 94, HHB 117, HHB 216	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
Ligule hair	Present	24	HHB 67 Improved, HHB 197, HHB 223, HHB 226, HHB 94, HHB 117, HHB 216	ICMA 843-22, ICMB 843-22, ICMA 97111, ICMB 97111, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
Seed covering	Exposed	13	HHB 67 Improved, HHB 223, HHB 226, HHB 94, HHB 117, HHB 216	ICMA 97111, ICMB 97111, HBL 11, H 77/833-2-202, H 77/29-2, G 73-107, HTP 3/13
	Intermediate	11	HHB 197	ICMA 843-22, ICMB 843-22, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B

Table 2. Contd.

Grain yield (g/plant)	Low (<15)	2	-	HMS 7A, HMS 7B
	Medium (16-30)	12	-	ICMA 843-22, ICMB 843-22, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, HMS 7A, HMS 7B, HBL 11, H 77/833-2-202, G 73-107, HTP 3/13
	High (31-50)	4	HHB 67 Improved	ICMA 97111, ICMB 97111, H 77/29-2
	Very high (>50)	6	HHB 223, HHB 226, HHB 94, HHB 117, HHB 216 HHB 197	-
Fresh fodder weight (g/plant)	Low (<200)	4	-	HMS 37A, HMS 37B, HMS 7A, HMS 7B
	Medium (201-300)	7	-	ICMA 843-22, ICMB 843-22, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, H 77/833-2-202,
	High (301-500)	11	HHB 67 Improved HHB 223, HHB 226, HHB 117, HHB 197	ICMA 97111, ICMB 97111, HBL 11, G 73-107, HTP 3/13, H 77/29-2
	Very high (>500)	2	HHB 94, HHB 216	-
Dry fodder weight (g/plant)	Low (<100)	2	-	HMS 7A, HMS 7B
	Medium (101-200)	12	-	ICMA 843-22, ICMB 843-22, ICMA 89111, ICMB 89111, ICMA 94555, ICMB 94555, HMS 37A, HMS 37B, H 77/833-2-202, H 77/29-2, G 73-107, HBL 11
	High (201-300)	7	HHB 67 Improved HHB 223, HHB 226, HHB 197	ICMA 97111, ICMB 97111, HTP 3/13
	Very high (>300)	2	HHB 94, HHB 216, HHB 117	-

their male parent HBL 11 and HTP 3/13 had brown bristles. Spike shape classified all the genotypes into four categories: conical, cylindrical, candle and lanceolate. H 77/833-2-202 had lanceolate spike shape, five genotype showed candle spike shape, another five showed cylindrical spike shape and the remaining thirteen were conical in spike shape. Spike tip sterility was present in sixteen genotypes and absent in the remaining eight genotypes. Nine genotypes showed compact spike density, four showed semi compact and the remaining eleven genotypes had loose spike.

Information outlined in Table 2 can effectively be used to find out distinct features of pearl millet genotypes. For example, HHB 117 was the only genotype having brown spikelet glum colour. The HMS 7A and HMS 7B were the only genotype having sheath pubescence. The HHB 94, ICMB 97111 and G 73-107 had brown anther colour. Whitish nodal pigmentation is the unique feature of G 73-107 and purple nodal pigmentation is the unique feature of ICMA 97111 and ICMB 97111. It is also found that partial ear head exertion type is

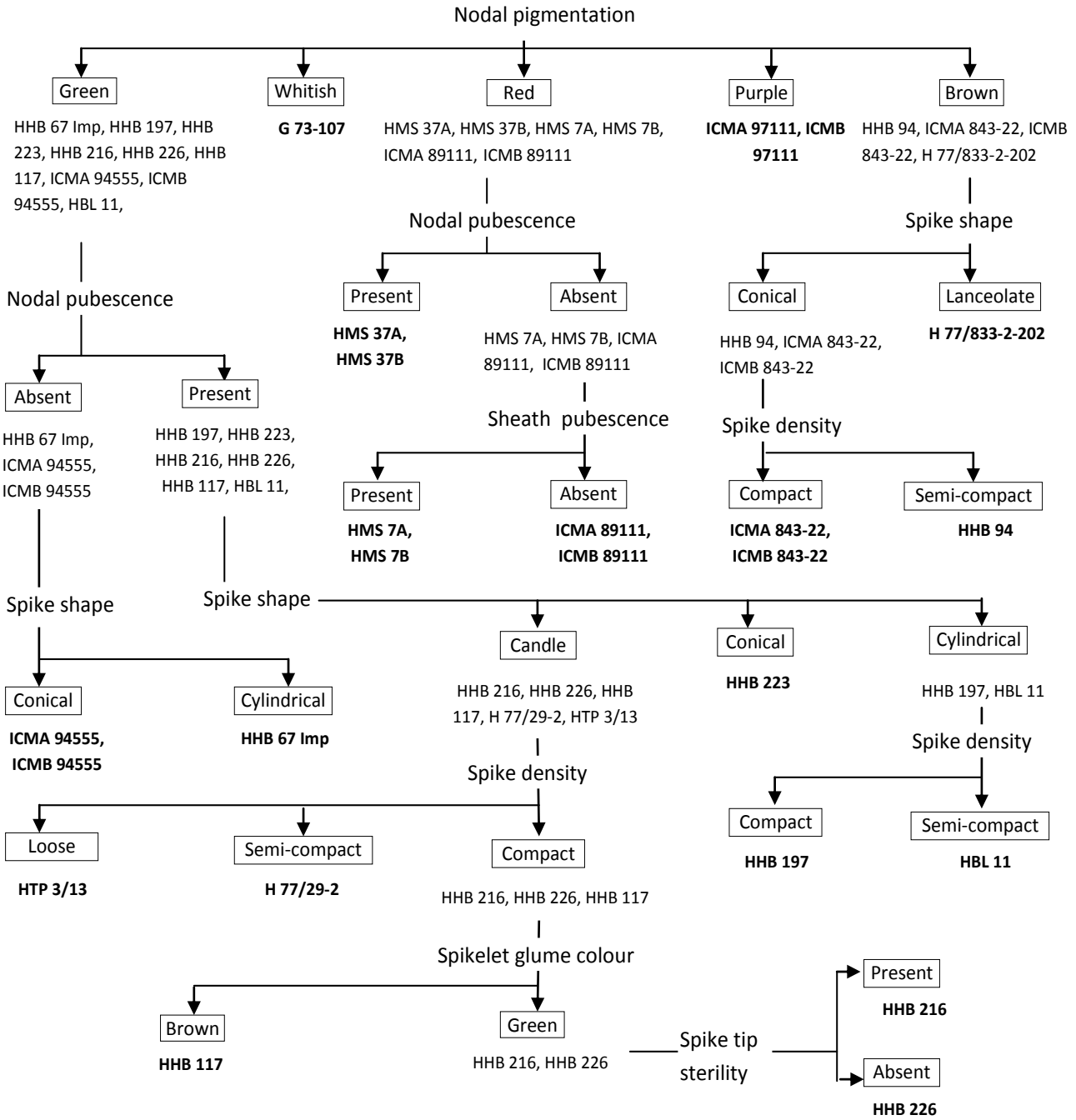

Figure 1. Flow chart diagram based on qualitative characters to identify the key diagnostic characters of pearl millet genotypes.

only found in HMS 37A and HMS 37B. H 77/833-2-202 was the only genotype having lanceolate spike shape. These are the qualitative characters and they can help in clear cut identification of particular genotype (Gupta et al., 2011).

Observations on plant height (short, medium, tall, very tall), leaf length (medium, long, very long), leaf width (narrow, medium, broad), number of nodes (low, medium), spike length (small, intermediate), bristle length (short, medium), grain yield/plant (low, medium, high, very high), fresh fodder weight/plant and dry fodder weight/plant (low, medium, high, very high) were recorded and the data was pooled over the locations. These could be grouped into distinct classes and could be useful for varietal identification and genetic purity testing.

In the Figure 1, flow chart was prepared using qualitative characters to find out distinctive features of all

the genotypes (Rana, 2004). Nodal pigmentation was considered as primary character keeping in view the early identification of genotype during plant growth. Nodal pigmentation classified all the genotypes into five groups green, whitish, red, purple and brown. The restorer line G 73-107 could easily be distinguished at early stage of crop growth by having whitish nodal pigmentation. The ICMA 97111 and ICMB 97111 could easily be distinguished by purple nodal pigmentation. Other three nodal pigmentation classes were further separated using nodal pubescence and spike shape. At this level H 77/833-2-202 could easily be distinguished by combination of two characters, that is, nodal pigmentation (brown) and spike shape (lanceolate). In the same way, distinguishing features of all the 24 genotypes were identified through this flow chart based on seven qualitative characters (nodal pigmentation, nodal pubescence, spike shape, spike density, spikelet glume colour, spike tip sterility and sheath pubescence). For example, HHB 94 having a path of three characters spike shape (conical), spike density (semi compact) and nodal pigmentation (brown) is unique for this genotype. It means no other genotype can follow or repeat this path, so this unique path revealed the key diagnostic features of HHB 94. In this flow chart, the CMS line and maintainer line have similar pathway because these two lines are isogenic lines. For example, HMS 7A and HMS 7B have the same key diagnostic features and common path in flow charts.

It may also be concluded that majority of the qualitative/morphological characters found to be dominant in the hybrids were contributed by the male parent. Bristles (present/absent), nodal pubescence, seed covering were found as dominant characters. The study revealed that the morphological characters studied (Table 2), can be used to characterize and catalogued pearl millet genotypes. The key diagnostic characters assigned to the genotypes may also be used for maintaining their seed lots and verification of identity and assessment of varietal purity for seed production, certification and quality control.

Conflict of Interests

The authors have not declared any conflict of interests.

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Vegetation responses to site, elevation and land use in semi-arid rangeland of Southern Ethiopia

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The dynamics of rangeland vegetation are products of spatial and temporal land use that determine rangeland productivity and conservation of biodiversity. This study examined effects of site, elevation, land use and season on vegetation attributes at three sites in Dida-Hara, southern Ethiopia. Herbaceous plant attributes (that is, above-ground herbaceous biomass, basal cover, plants' density, species richness, diversity index and evenness), as well as woody plants characteristics such as density and species richness were measured. Herbaceous and woody vegetation variables were examined by season and land use types (that is, enclosures vs. open-grazed areas) across three sites and two altitudes. The results showed that all vegetation attributes were greatly affected by site, land use type and season. Herbaceous vegetation attributes such as biomass, basal cover and herbaceous species richness were more affected by land use types and season. Elevation affected herbaceous vegetation characteristics such as basal cover, herbaceous species diversity and woody richness. Effects of site differences in terms of herbaceous biomass were common during the dry season. Grass diversity was significantly affected by site, elevation, land use type, season and altitude across spatial and temporal scales. Herbaceous biomass was significantly higher in enclosure than in the communal land use type whereas herbaceous biomass showed a declining trend with increased density of woody plants.

Key words: Vegetation attributes, rangeland, grazing pressure, elevation, site, land-use, season.

INTRODUCTION

Knowledge of vegetation dynamics at different spatial and temporal scales in relation to different land use practices, elevation and season is essential to consider for proper management of arid rangelands (Fernandez-Gimenez and Allen-Diaz, 1999; Snyder and Tartowski, 2006). This is particularly important in arid and semi-arid rangelands where forage productivity is highly variable across sites, between seasons and altitudinal gradient (Gibbens et al., 2005; Oba et al., 2000; Briske et al., 2003; Peters and Havstad, 2006). We need to isolate the natural drivers of rangeland vegetation dynamics from human induced impacts (Fernandez-Gimenez and Allen-Diaz, 1999; Bestelmeyer et al., 2006). Among the drivers of rangeland vegetation dynamics, land use practices

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(Coppock, 1994; Angassa and Oba, 2010), fire suppression (Oba et al., 2000; Angassa and Oba, 2008), site differences and altitudinal variation (Sheuyange et al., 2005; Wu and Archer, 2005; Bestelmeyer, et al., 2006; Dime et al., 2012) and impact of recurrent drought (Peters and Havstad, 2006) can affect the dynamics of rangeland vegetation.

Peters and Havstad (2006) have shown that the spatial distribution of rangeland vegetation across site can be greatly affected by the physical environments such as soil differences, amount and distribution of rainfall. Others (Angassa and Oba, 2010; Teka et al., 2012; Angassa, 2014) indicated that season and land use types have a significant effect on the dynamics of rangeland vegetation. Management of grazing lands thus requires resource planners to be aware of the effects of site differences and land use types on rangeland productivity (Peters and Havstad, 2006; Riginos and Grace, 2008). Wu and Archer (2005) have argued that the extent to which variation among sites may favorably maintain soil moisture and nutrients can be a crucial element of vegetation dynamics. Hence, patterns of vegetation dynamics on rangeland ecological sites can be expressed in terms of climate variability, soil types, soil moisture and nutrient availability (Briske et al., 2003). Although a number of factors might be responsible for the dynamics of rangeland vegetation, the combined effects of site, elevation and land use in relation to season have remained unclear. Thus, how site differences, altitudinal gradient and land use types influence vegetation dynamics on rangelands needs better understanding (Oba et al., 2000; Angassa, 2005).

In the case of arid and semi-arid rangelands, one would envisage, the temporal variability being critically important for herders as they use grazing lands in rotation based on seasonal mobility. This means that the temporal variation acts on the spatial variability of grazing lands to influence the dynamics of land use patterns between seasons of the year (Fernandez-Gimenez and Allen-Diaz, 1999; Bestelmeyer et al., 2006; Snyder and Tartowski, 2006). How gazing or lack of disturbance by livestock influences the products of the interactions between spatial and temporal scales in arid and semi-arid rangelands is important to note. For example, do we have the same outcome as the communal rangelands if livestock grazing is excluded for longer period from a given rangeland site? What would be the responses of vegetation variables if grazing pressure is removed on the one hand, and continuous grazing is allowed on adjacent rangeland areas? Understanding the effects of site, altitudinal gradient and land use types has important implications for proper management and conservation of rangeland biodiversity.

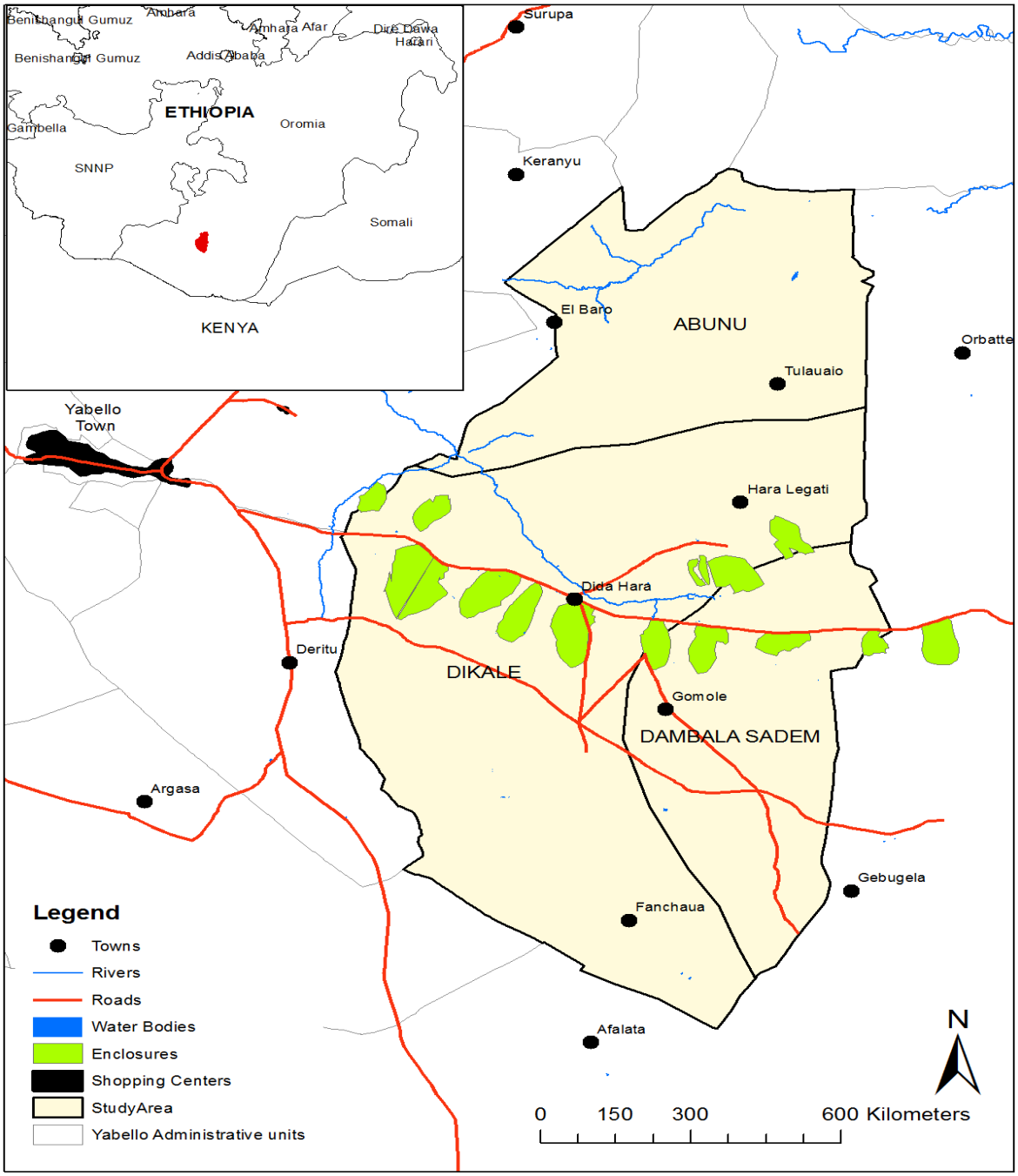
The present study evaluates responses of vegetation attributes to site, altitudinal variation and land use types in Borana rangelands of southern Ethiopia. To achieve the objective of this study, the following questions were asked: (1) How do spatial and temporal variability of rangelands influence herbaceous vegetation attributes (that is, herbaceous biomass, basal cover, density, species richness, diversity and evenness) and woody vegetation variables (that is, density and richness)? (2) What is the effect of land use types, site differences, altitudinal gradient and season on vegetation variables?
It was predicted that the impacts of spatial scales are more positive in facilitating woody vegetation than human induced land use types, while the temporal scales influence herbaceous biomass production, keeping the land use type constant. It was also predicted that the interactions between season and grazing pressure would show greater influence on herbaceous vegetation than on woody vegetation attributes.

MATERIALS AND METHODS

Description of the study sites

This study was conducted in semi-arid rangelands of southern Ethiopia across three locations (Dikale, Dembi and Siiquu) in Dida-Hara (Picture 1). The study locations hereafter referred to as sites based on two types of land use systems (enclosures vs. communal land use) where cattle are the dominant livestock type. Mean annual rainfall from the nearest weather stations over the last two decades was 500 mm (Angassa and Oba, 2007). Soil types of the three study sites vary from red in the uplands to ‘‘black cotton’’ soils in the bottomlands as in all the three sites, land use types were replicated twice by altitudinal gradient for data collection (Coppock, 1994). *Commiphora africana*, *Acacia drepanolobium* and other *Acacia* species dominate the woody cover, while grass species such as *Chrysopogon aucheri, Cenchrus ciliaris, Aristida adscensionis, Eragrostis papposa, Heteropogon contortus, Panicum coloratum, Pennisetum mezianum, Pennisetum stramineum* and *Themeda triandra* make up the majority of herbaceous vegetation in the study sites (Angassa and Baars, 2000).

The study questions were examined by conducting vegetation sampling at different altitudes in relation to season and grazing pressure to understand their effects on rangeland vegetation dynamics. According to Peters and Havstad (2006), plot is a patchy spatial unit in association with plant communities. In vegetation samplings, plots are the smallest sampling units nested within landscape patches. The same authors stated that patchy plant communities are often associated with unique microclimate, variation in the physical environments and weather conditions interacting with vegetation community to influence variations among ecological units that also affect plant community success with consequences for rangeland vegetation patterns. Vegetation responses to rangeland ecological variables at broader spatial scales can vary from those influences at intermediate and fine scales (Peters and Havstad, 2006). The second scale of interest in this study was the altitudinal gradient, consisting of groups of patchy mosaics of rangeland units. The scale for the altitude and plot matrixes was assessed based on uplands and bottomlands. The third scale used for the evaluation of rangeland vegetation dynamics was land use types or grazing pressure (that is, enclosures vs. communal rangelands) replicated across two elevation ranges. The fourth independent variable was season of rainfall as a factor regulating plant productivity on the rangelands.



Picture 1. Map of the study area in Dida-Hara, southern Ethiopia.

Sampling procedure

In each study location, two land-use types (that is, enclosures vs. communal grazing lands) were identified. Enclosures are areas protected from livestock grazing to conserve pasture for calves for dry season grazing, while communal grazing lands are continuously grazed areas throughout the year and lacks rest for plant species to recover. These land-use types were replicated twice in both upland and bottom land altitudes for sampling vegetation in enclosures vs. communal rangelands by altitude. Upland altitudes are areas > 1500 m above sea level (m.a.s.l.), while area < 1500 m.a.s.l was considered as bottomlands. A 500 m long transect was established at each site in each land-use system per altitude gradient. The 500 m long transect was divided into 10 units consisting of 10 plots at 50 m intervals between each plot. Vegetation variables were sampled in each plot along the transect within each land-use

system per altitude using 1 m × 1 m plot to quantify herbaceous vegetation, whereas 10 m × 10 m plot was used for sampling woody plants. During two rainy seasons and one dry season (that is, in May and November, 2004, and February, 2005, respectively), biomass, basal cover, species richness, diversity and evenness of herbaceous plants, as well as density and richness of woody plants were measured. Plant species were identified and counted in each plot. Herbaceous biomass was harvested using hand cutting and samples were oven dried at 65°C for 24 h. Herbaceous biomass was determined based on the dry matter estimation of herbaceous samples. The proportion of grass basal cover was estimated visually based on the area (soil part) covered by a grass base compared with bare ground (Angassa and Oba, 2010). The density of woody species were determined by counting the total number of individuals in plots of 10 m × 10 m. Species richness were counted per plot per census (that is, the total count of all species per plot), while species diversity index, $H' = -(\Sigma (p_i \ln p_i))$ (Shannon, 1948) was calculated using the relative abundance of each plant species (where pi represents the proportion of individual species in each plot).

Data analysis

SAS statistical software version 9.1 (SAS Institute, 2001) was used for statistical analysis. Analysis of variance (ANOVA) was used to evaluate vegetation attributes (that is, biomass, basal cover, density, species richness, diversity index and evenness of herbaceous plants) and woody vegetation variables (woody density and richness) in response to spatial scales in rangeland ecological units. Post-hoc comparisons of significant differences were done using the least square difference (LSD) method. The relationship between woody density and grass biomass were also determined using a polynomial regression. Statistical significance was reported at $P< 0.05$.

RESULTS

Site, elevation and land use types on vegetation attributes

The present results showed that all vegetation variables were significantly related to site ($P < 0.05$) (Table 1). Season and land use types were significantly related to most herbaceous vegetation variables ($P < 0.001$). Interactions between site and season, as well as season and management (that is, land use types) on the yield of grasses were significant ($P < 0.001$) (Table 1). Grass basal cover, herbaceous diversity and woody richness were significantly related to the effects of altitudinal gradient (Table 1). There were significant interactions between site and altitude on grass basal cover, herbaceous species richness and diversity, and woody richness (Table 1). The interaction between season and altitude was significantly related to woody richness (Table 1).

Season

No significant variation in grass yield was recorded between the study sites and interactions between site and season (Table 2). However, site was significantly ($P < 0.05$) related to the yield of grasses during the dry season (Tables 2 and 3). The interaction between site and altitude also significantly ($P < 0.05$) affected the basal cover of grasses during the short rainy season (Table 2). Grass density was significantly ($P < 0.05$) differed between sites despite seasonal variation (Table 2). Likewise, grass density was significantly ($P < 0.05$) affected by the interaction between site and altitude during the different seasons (Table 2). Site had a significant effect on grass richness only during the short and dry seasons. Grass diversity was significantly affected at all spatial scales and their interaction (exception being the interaction between site and plot) during the short rainy season (Table 2). Hence, our results showed that the interaction between site and landscape had a significant effect on grass diversity during the main rainy season (Table 2). Moreover, grass diversity was significantly affected by site and landscape, as well as the interaction between site and landscape during the dry season (Tables 2 and 4). Woody plant density was significantly affected by site, landscape and their interaction during the main and short rainy seasons (Tables 2 and 3). Similarly, the interaction between site and plot had a significant effect on the density of woody plants during the short rainy season (Table 2). Woody plant richness was significantly affected by site and the interaction between site and landscape during the main rainy season. Overall, site and landscape had a significant effect on woody plant richness during the short rainy and dry seasons.

Land-use

Grass biomass was significantly ($P < 0.05$) affected by site differences under the enclosure (Tables 5 and 6). However, the interaction effects were non-significant ($P > 0.05$) at altitude and plot levels when considering the yield of grasses under both land-use systems (Table 5). The basal cover of grasses showed significant differences both at site and altitude levels under enclosure management (Tables 5 and 7). The basal cover of grasses was significantly affected by site, and the interaction between site and altitude under communal land-use (Table 5). Site differences had a significant effect on the density of grasses under both land-use systems, while the interaction between site and landscape was only significant under the communal land-use (Table 5). Site differences significantly affected grass richness under both land-use systems (Tables 5 and 6). Nevertheless, the effect of altitude on grass richness was only significant under the communal land-use. The interaction between site and altitude had a significant effect on grass richness under both land-use systems.

Table 1. Main and interaction effects of site, elevation, season and land use types (enclosure vs. communal land use) on vegetation attributes as affected in Borana, southern Ethiopia.

Dependent variables	Independent variable	Df	F	P
Grass biomass (gm m^{-2})	Site	2	4.26	0.0149
	Altitude	1	0.04	0.8423
	Season	2	124.21	<0.0001
	Management	1	156.59	<0.0001
	Site*altitude	2	0.06	0.9376
	Site*Season	4	6.07	<0.0001
	Site*Management	2	2.09	0.1255
	Altitude*Season	2	2.76	0.0647
	Altitude*Management	1	0.01	0.9210
	Season*Management	2	25.75	<0.0001
Basal cover(%)	Site	2	5.73	0.0036
	Altitude	1	5.71	0.0174
	Season	2	12.76	<0.0001
	Management	1	145.26	<0.0001
	Site*altitude	2	5.51	0.0044
	Site*Season	4	1.14	0.3395
	Site*Management	2	5.00	0.0072
	Altitude*Season	2	0.31	0.7303
	Altitude*Management	1	1.94	0.1641
	Season*Management	2	5.53	0.0043
Grass richness (No. m^{-2})	Site	2	12.13	<0.0001
	Altitude	1	2.60	0.1080
	Season	2	3.86	0.0221
	Management	1	20.43	<0.0001
	Site*altitude	2	12.99	<0.0001
	Site*Season	4	4.24	0.0023
	Site*Management	2	1.47	0.2323
	Altitude*Season	2	0.78	0.4571
	Altitude*Management	1	2.60	0.1080
	Season*Management	2	0.47	0.6257
Grass diversity (H´)	Site	2	6.53	0.0017
	Altitude	1	20.81	<0.0001
	Season	2	16.98	<0.0001
	Management	1	14.45	0.0002
	Site*altitude	2	10.18	<0.0001
	Site*Season	4	0.53	0.1713
	Site*Management	2	0.48	0.6187
	Altitude*Season	2	0.98	0.3825
	Altitude*Management	1	0.04	0.8388
	Season*Management	2	3.67	0.0265
Woody density (stem/ha)	Site	2	7.32	0.0008
	Altitude	1	2.18	0.1408
	Season	2	5.47	0.0046
	Management	1	1.17	0.2802

Table 1. Contd.

	Site*altitude	2	1.73	0.1784
	Site*Season	4	4,22	0.0024
	Site*Management	2	1.65	0.1927
	Altitude*Season	2	2.57	0.0777
	Altitude*Management	1	1.24	0.2670
	Season*Management	2	1.44	0.2392
Woody richness (100 m^{-2})	Site	2	21.40	<0.0001
	Altitude	1	49.09	<0.0001
	Season	2	8.33	0.0003
	Management	1	0,04	0.8499
	Site*altitude	2	3.62	0.0277
	Site*Season	4	2.47	0.0447
	Site*Management	2	0.41	0.6610
	Altitude*Season	2	6.91	0.0011
	Altitude*Management	1	0.04	0.8499
	Season*Management	2	5.75	0.0035

Table 2. Analysis of variance related to site, altitude and plot level effects on vegetation dynamics over three seasons in southern Ethiopia.

Dependent variable	Independent variable	Df	Main rain		Short rain		Dry season	
			F	*P*	F	*P*	F	*P*
Grass yield (gm m^{-2})	Site	2	2.68	0.0744	1.32	0.2714	6.34	0.0027
	Atitude	1	1.18	0.2797	0.38	0.5383	1.84	0.1785
	Plot	9	1.41	0.1943	0.30	0.9725	0.29	0.9750
	Site*altitude	2	0.08	0.9276	0.11	0.8942	0.23	0.7920
	Site*plot	18	0.67	0.8338	0.26	0.9989	0.62	0.8725
Basal cover (%)	Site	2	1.61	0.2049	1.19	0.3104	2.49	0.0890
	Altitude	1	1.62	0.2071	1.47	0.2284	0.48	0.4890
	Plot	9	0.45	0.9047	0.65	0.7534	0.37	0.9453
	Site*altitude	2	0.26	0.7699	3.19	0.0459	0.82	0.4440
	Site*plot	18	0.39	0.9861	0.82	0.6756	0.39	0.9869
Grass density (stem m^{-2})	Site	2	4.35	0.0158	5.47	0.0058	6.92	0.0016
	Altitude	1	0.01	0.9044	0.56	0.4553	0.02	0.8763
	Plot	9	1.99	0.0500	1.73	0.0952	0.37	0.9445
	Site*altitude	2	4.88	0.0098	6.31	0.0028	7.49	0.0010
	Site*plot	18	1.03	0.4379	1.00	0.4687	0.75	0.7524
Grass richness (No. m^{-2})	Site	2	0.74	0.4790	7.31	0.0012	15.18	<0.0001
	Altitude	1	0.00	0.9546	0.93	0.3384	3.84	0.0534
	Plot	9	0.88	0.5433	0.32	0.9673	1.06	0.3969
	Site*altitude	2	1.19	0.3108	8.76	0.0003	8.02	0.0006
	Site*plot	18	1.05	0.4126	1.13	0.3428	1.58	0.0826
Grass diversity (H´)	Site	2	1.01	0.3673	3.89	0.0241	3.28	0.0421
	Altitude	1	3.09	0.0823	4.99	0.0280	17.12	<0.0001

Table 2. Contd.

	Plot	9	1.04	0.4139	2.24	0.0264	1.42	0.1919
	Site*altitude	2	5.39	0.0062	5.33	0.0066	2.29	0.1072
	Site*plot	18	0.93	0.5467	0.98	0.4896	0.82	0.6701
	Site	2	11.13	<.0001	117.52	<.0001	0.17	0.8450
	Altitude	1	20.43	<.0001	17.55	<.0001	0.36	0.5506
Woody density (stem/ha)	Plot	9	0.85	0.5722	1.74	0.0928	0.97	0.4723
	Site*altitude	2	9.88	0.0001	28.84	<.0001	2.04	0.1368
	Site*plot	18	0.92	0.5552	1.79	0.0390	1.12	0.3515
	Site	2	8.49	0.0004	5.87	0.0040	11.41	<0.0001
	Altitude	1	3.28	0.0735	11.00	0.0013	47.42	<0.0001
Woody richness (100 m^{-2})	Plot	9	1.02	0.4339	1.18	0.3185	0.93	0.5033
	Site*altitude	2	7.85	0.0007	1.69	0.1910	1.03	0.3614
	Site*plot	18	0.97	0.5026	0.58	0.9061	0.77	0.7318

Table 3. Site dependent effects on rangeland vegetation responses over three seasons in southern Ethiopia.

Response variable	Site	Main rain	Short rain	Dry season
	Dikale	260 ± 9.4^{a}	124 ± 10.2^{b}	128 ± 8.22^{b}
Grass yield (gm m^{-2})	Dambi	228 ± 9.4^{b}	163 ± 10.2^{a}	153 ± 8.22^{a}
	Siiquu	241 ± 9.4^{ab}	141 ± 10.2^{ab}	91 ± 8.41^{c}
	Dikale	16.4 ± 0.75^{a}	17 ± 0.98	13 ± 0.83^{ab}
Basal cover (%)	Dambi	13.8 ± 0.75^{b}	15 ± 0.98	11 ± 0.83^{b}
	Siiquu	14.4 ± 0.75^{ab}	16 ± 0.98	14 ± 0.83^{a}
	Dikale	41 ± 1.68^{a}	65 ± 2.84^{a}	46 ± 2.11^{a}
Grass density (stem m^{-2})	Dambi	34 ± 1.68^{b}	51 ± 2.84^{b}	36 ± 2.11^{b}
	Siiquu	38 ± 1.68^{ab}	55 ± 2.84^{b}	36 ± 2.11^{b}
	Dikale	5.1 ± 0.25	4 ± 0.17^{b}	4.4 ± 0.21^{b}
Grass richness (No. m^{-2})	Dambi	5.2 ± 0.25	5 ± 0.17^{a}	5.6 ± 0.21^{a}
	Siiquu	4.8 ± 0.25	5 ± 0.17^{a}	4.1 ± 0.21^{b}
	Dikale	2.0 ± 0.13	2.2 ± 0.11^{b}	1.7 ± 0.10^{b}
Grass diversity (H´)	Dambi	1.8 ± 0.13	2.2 ± 0.11^{b}	1.7 ± 0.10^{b}
	Siiquu	2.0 ± 0.13	2.6 ± 0.11^{a}	2.0 ± 0.10^{a}
	Dikale	0.60 ± 0.04	0.67 ± 0.03^{b}	0.52 ± 0.03^{b}
Grass evenness	Dambi	0.55 ± 0.04	0.68 ± 0.03^{b}	0.52 ± 0.03^{b}
	Siiquu	0.63 ± 0.04	0.78 ± 0.03^{a}	0.62 ± 0.03^{a}
	Dikale	3515 ± 310^{b}	1930 ± 239.8^{b}	6165 ± 1162
Woody density (stem/ha)	Dambi	5480 ± 310^{a}	6771 ± 239.8^{a}	5243 ± 1162
	Siiquu	3450 ± 310^{b}	2500 ± 239.8^{b}	5470 ± 1162
	Dikale	4.5 ± 0.24^{a}	3.9 ± 0.22^{a}	4.7 ± 0.22^{a}
Woody richness (100 m^{-2})	Dambi	4.4 ± 0.24^{a}	3.4 ± 0.22^{a}	3.4 ± 0.22^{b}
	Siiquu	3.3 ± 0.24^{b}	2.8 ± 0.22^{b}	3.4 ± 0.22^{b}

a, b and c = superscripts with different letters within columns showing significant differences among sites within seasons.

Table 4. Altitude dependent effects on vegetation variables over three seasons in southern Ethiopia.

Response variable	Altitude	Main rain	Short rain	Dry season
Grass yield (gm m^{-2})	Upland	249±8.16	148±12.59	113±10.18
	Bottomland	236±8.16	136±12.59	134±10.18
Basal cover (%)	Upland	140.80	15±1.00	12±0.68
	Bottomland	16±0.80	17±1.00	13±0.68
Grass density (stem m^{-2})	Upland	37±1.50	55±2.74	40±1.84
	Bottomland	38±1.50	58±2.74	39±1.84
Grass richness (No. m^{-2})	Upland	5.01±0.20	4.5±0.18	4.5±0.20
	Bottomland	5.03±0.20	4.7±0.18	4.9±0.20
Grass diversity (H´)	Upland	2.1±0.11	2.5±0.10[a]	2.05±0.09
	Bottomland	1.8±0.11	2.2±0.10[b]	1.57±0.09
Grass evenness	Upland	0.63±0.03	0.75±0.03	0.63±0.03
	Bottomland	0.55±0.03	0.67±0.03	0.48±0.03
Woody density (stem/ha)	Upland	5051±322	4324±379	5221±964
	Bottomland	3246±322	3143±379	6031±964
Woody richness (100 m^{-2})	Upland	3.82±0.20	2.9±0.19	2.90±0.19
	Bottomland	4.28±0.20	3.75±0.19	4.67±0.19

Table 5. Analysis of variance related to site and altitude on vegetation dynamics under different land-use systems in southern Ethiopia.

Response variable	Independent variable	DF	Enclosure		Communal	
			F	*P*	F	*P*
Grass yield (gm m^{-2})	Site	2	3.85	0.0242	0.36	0.6999
	Altitude	1	0.01	0.9301	0.00	0.9633
	Plot	9	0.89	0.5328	0.24	0.9885
	Site*altitude	2	0.20	0.8228	0.12	0.8846
	Site*plot	18	0.42	0.9819	0.22	0.9997
Basal cover (%)	Site	2	3.30	0.0406	9.38	0.0002
	Altitude	1	5.42	0.0213	0.68	0.4095
	Plot	9	1.48	0.1627	1.10	0.3642
	Site*altitude	2	2.21	0.1133	4.81	0.0095
	Site*plot	18	1.10	0.3623	1.50	0.0975
Grass density (stem m^{-2})	Site	2	4.62	0.0114	7.58	0.0008
	Altitude	1	3.23	0.0746	0.87	0.3533
	Plot	9	1.16	0.3246	1.06	0.3951
	Site*altitude	2	2.11	0.1246	15.33	<0.0001
	Site*plot	18	0.77	0.7335	0.61	0.8835
Grass richness (No. m^{-2})	Site	2	7.54	0.0008	6.32	0.0024

Table 5. Contd.

	Altitude	1	0.00	1.0000	5.61	0.0192
	Plot	9	0.84	0.5777	1.06	0.3984
	Site*altitude	2	6.20	0.0026	11.08	<0.0001
	Site*plot	18	1.16	0.3005	1.64	0.0591
	Site	2	3.13	0.0468	3.15	0.0458
	Altitude	1	9.25	0.0028	9.50	0.0025
Diversity index (H´)	Plot	9	0.71	0.6960	1.51	0.1514
	Site*altitude	2	4.31	0.0153	5.33	0.0059
	Site*plot	18	0.89	0.5939	0.97	0.4983
	Site	2	3.49	0.0477	18.28	<.0001
	Altitude	1	1.73	0.1909	0.27	0.6021
Woody density (stem/ha)	Plot	9	0.74	0.6759	0.98	0.4569
	Site*altitude	2	0.03	0.9724	13.31	<0.0001
	Site*plot	18	0.92	0.5343	1.21	0.2641
	Site	2	15.21	<0.0001	11.62	<0.0001
	Altitude	1	29.29	<0.0001	32.25	<0.0001
Woody richness(100 m^{-2})	Plot	9	0.89	0.5319	5.54	<0.0001
	Site*altitude	2	7.40	0.0009	0.04	0.9648
	Site*plot	18	4.04	<0.0001	2.19	0.0008

Table 6. Site dependent effects on responses of rangeland vegetation under different land-use systems in southern Ethiopia.

Response variable	Site	Enclosure	Communal
	Dikale	207±10.97^{ab}	135±11.43
Grass biomass (gm m^{-2})	Dambi	232±10.97^{a}	130±11.43
	Siiquu	195±10.97^{b}	120±11.43
	Dikale	19.3±0.75^{a}	12±0.62^{a}
Basal cover (%)	Dambi	17.2±0.75^{b}	9±0.62^{b}
	Siiquu	16.7±0.75^{b}	13±0.62^{a}
	Dikale	49.6±1.59^{a}	39±2.55^{b}
Grass density(stem m^{-2})	Dambi	42.1±1.59^{b}	52±2.55^{a}
	Siiquu	42.1±1.59^{b}	44±2.55^{b}
	Dikale	4.7±0.18^{b}	4.33±0.18^{b}
Grass richness (No. m^{-2})	Dambi	5.6±0.18^{a}	4.90±0.18^{a}
	Siiquu	5.0±0.18^{b}	4.08±0.18^{b}
	Dikale	2.13±0.10^{b}	1.76±0.10^{b}
Grass diversity (H´)	Dambi	1.99±0.10^{b}	1.81±0.10^{b}
	Siiquu	2.35±0.10^{a}	2.06±0.10^{a}
	Dikale	0.65±0.03^{b}	0.54±0.03^{b}
Grass evenness	Dambi	0.61±0.03^{b}	0.55±0.03^{b}

Table 6. Contd.

	Siiquu	0.72±0.03[a]	0.63±0.03[a]
Woody density (stem/ha)	Dikale	3576±822[b]	4164±324[b]
	Dambi	6122±822[a]	5540±324[a]
	Siiquu	4607±822[ab]	3007±324[c]
Woody richness (100 m^{-2})	Dikale	4.4±0.20[a]	4.3±0.18[a]
	Dambi	3.7±0.20[b]	3.7±0.18[b]
	Siiquu	3.0±0.20[c]	3.3±0.18[c]

Table 7. Altitude dependent effects on responses of rangeland vegetation under different land-use systems in southern Ethiopia.

Response variable	Altitude	Enclosure	Communal
Grass yield g/m^2	Upland	212±9.10	128.4±9.30
	Bottomland	211±9.10	127.7±9.30
Basal cover (%)	Upland	17±0.67[b]	10.7±0.53
	Bottomland	19±0.67[a]	11.3±0.53
Grass density (stem m^{-2})	Upland	43±1.68	46±2.14
	Bottomland	47±1.68	43±2.14
Grass richness (No. m^{-2})	Upland	5.07±0.16	4.21±0.15[b]
	Bottomland	5.07±0.16	4.67±0.15[a]
Grass diversity (H´)	Upland	2.33±0.08[a]	2.04±0.08[a]
	Bottomland	1.98±0.08[b]	1.71±0.08[b]
Grass evenness	Upland	0.72±0.03	0.63±0.02[a]
	Bottomland	0.61±0.03	0.53±0.02[b]
Woody density (stem/ha)	Upland	5404±676	4326±285
	Bottomland	4133±676	4147±285
Woody richness (100 m^{-2})	Upland	3.18±0.17[b]	3.23±0.16[b]
	Bottomland	4.23±0.17[a]	4.230.16[a]

The results showed that grass diversity was significantly affected by site, altitudinal variation and their interaction under both land-use systems. Woody plant density significantly varied which was caused by site differences under both land-use systems (Tables 5 and 6). The interaction between site and landscape under both land-use systems was also significantly ($P < 0.05$) different. The current result showed that site differences had a highly significant effect on the density of woody vegetation attributes under the communal land-use system. Woody plant richness was significantly ($P < 0.05$) affected by site, altitude and the interaction between site and plot under both land-use systems. Overall, woody vegetation richness was significantly affected by the interaction between site and altitude under the enclosure management (Table 5). The relationship between woody density and grass yield is shown in Figures 1, 2 and 3. The present results showed that woody plant density was inversely related to grass cover in all study sites (Figures 1, 2 and 3).

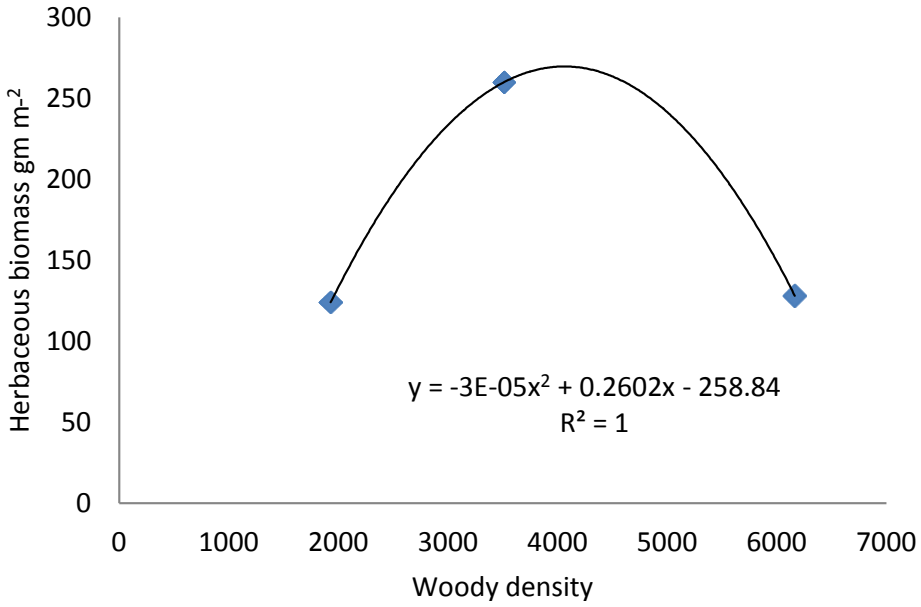


Figure 1. Relationship between grass yield and woody density in Dikale area, Borana.

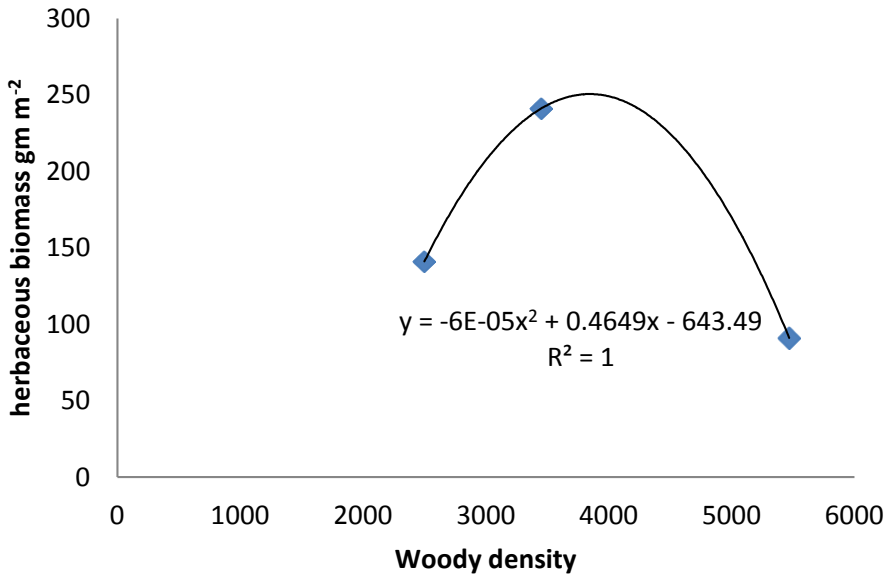


Figure 2. Relationship between grass yield and woody density in Siiquu area, Borana.

DISCUSSION

Effects of site, elevation and land use on vegetation attributes

It was predicted that the impacts of spatial scales are more decisive in shaping the distribution of vegetation than management intervention in arid rangelands, while the temporal scales could influence vegetation production, keeping land use type constant. Such impacts were observed in terms of woody vegetation composition, while herbaceous biomass was strictly controlled by grazing pressure and season. Oba et al. (2003) suggested that rangeland spatial variability would tend to increase differences in vegetation composition and production. The findings of the present results

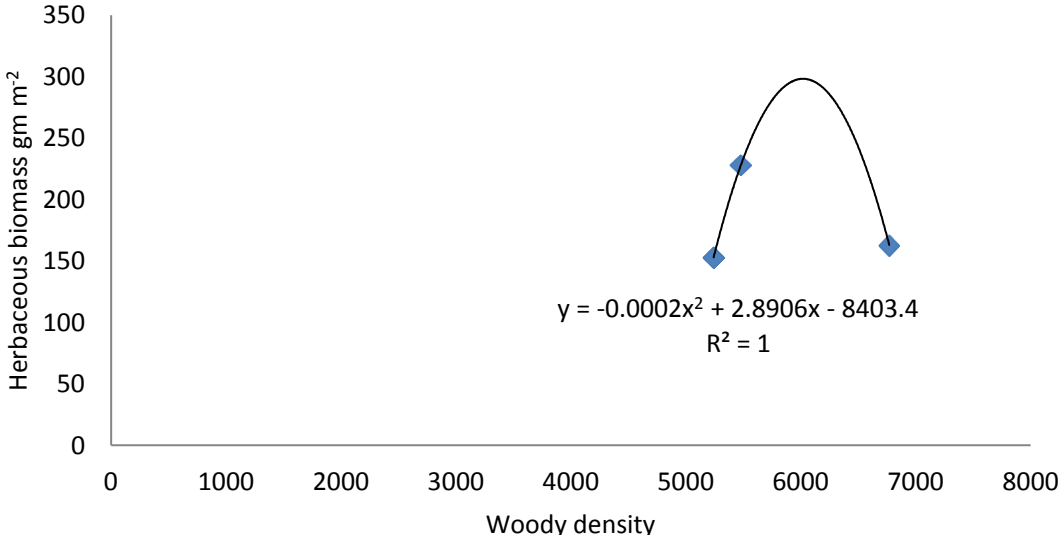


Figure 3. Relationship between grass yield and woody density in Denbi area, Borana.

indicate that vegetation responses could be influenced by the spatial and temporal variability of arid rangelands. Oba et al. (2000) have also indicated that landscape heterogeneity and temporal scales would contribute to the spatial variability of rangeland vegetation. The current results also support this argument, as the observed vegetation variables greatly varied across spatial and temporal scales. Similarly, several authors, for example, Meentemeyer and Box, (1987), Turner et al. (1989), Oba et al. (2003), Peters and Havstad (2006), Snyder and Tartowski (2006) and Wu and Archer (2005) have argued that vegetation dynamics are more responsive to site differences and altitudinal gradient than land use types.

The present study found that yield of grasses during the dry season varied between sites. The findings of the present study show that the interaction between site and landscape has important effect on basal cover of grasses during the short rainy season. Generally, the current findings suggest that variation in rangeland sites has greatly influenced the density of grasses regardless of seasonal variability. Vegetation composition and productivity within a region basically reflect the existing climate in terms of season and annual rainfall variability (Fernandez-Gimenez and Allen-Diaz, 1999; Oba et al., 2000; Peters and Havstad, 2006). Ultimately, climate variability plays a central role in determining the dynamics of rangeland vegetation (Ellis and Swift, 1988; Fernandez-Gimenez and Allen-Diaz, 1999; Briske et al., 2003). However, regional climatic variability may not account for the spatial patterns of vegetation at local level as a result of the irregularities that could arise due to altitudinal variation (Fernandez-Gimenezand and Allen-Diaz, 1999; Oba et al., 2000; Briske et al., 2003; Wu and Archer, 2005; Peters and Havstad, 2006). Hence, landscape spatial scales might exert a strong influence on the distribution, growth and abundance of plant communities over a wider altitudinal range due to substantial dissimilarities, in elevation, soil texture and moisture conditions, and nutrient supply (Wu and Archer, 2005). For example, Peters and Havstad (2006) have indicated that the nutrient and soil moisture distribution between high and low resource areas could be among the major factors for the variation in vegetation composition across spatial scales. Similarly, the results of the present study indicate that grass diversity was highly influenced by differences in site, altitude, season of rainfall. This is in accordance with the existing information that suggests that the dynamics of rangeland vegetation can be non-linear (Peters and Havstad, 2006) and locally mediated by the type of topography of an area (for example, Wu and Archer, 2005). These scale-dependent effects can have important implications for the local users and decision makers on the sustainability of arid land management.

Previous studies (for example, Ryerson and Parmenter, 2001; Briske et al., 2003) have shown that the occurrence of vegetation type along altitudinal gradient is independent of the exclusion of herbivores from a specific land site. The current study shows that grass density was greatly affected by the interaction between site and elevation during the different seasons, suggesting that plant composition always varies across landscapes. In addition to distinct and interactive effects of spatial scales, environmental variables such as season and land-use type are among the most important factors in driving vegetation dynamics (Peters and Havstad, 2006;

Briske et al., 2003). In some locations, grasses are diminished prior to drought, while in others grasses remain high regardless of the occurrence of drought (Peters and Havstad, 2006). Similarly, Coppock (1994) has reported that intensive grazing can lead to reduced grass cover and increased bush encroachment. On the other hand, exclusion of livestock grazing is found ineffective in limiting the spread of bush encroachment in semi-arid savannas (Peters and Havstad, 2006; Angassa and Oba, 2010).

According to Kerstin et al. (2005), increase in the density of woody encroachment suppresses grass production. From the results of the present study it seems that as the density of woody species increased grass biomass tends to reduce. In the present study, heavy grazing pressure is a common feature of the study sites.

Conclusion

It seems that the dynamics of rangeland vegetation that potentially accompany shifts in season or disturbance regimes is likely to be controlled by altitudinal gradient. Knowledge of vegetation dynamics may therefore need to explicitly account for the spatial and temporal variability of rangeland ecological sites in terms of season and land-use types to precisely predict the drivers of vegetation changes in savanna ecosystems.

Conflict of Interests

The authors have not declared any conflict of interests.

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Full Length Research Paper

Effect of planting axles, sunlight faces and rod vibration frequencies in the mechanized coffee harvesting

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Several studies have investigated the harvest systems, locations and conditions of the coffee crop, but there are no studies about the rod frequency vibration, particularly with regard to the effects of the position of the coffee with respect to exposure of sunlight regarding production, fruit maturation, and harvest levels. Thus, this study, conducted in two crops in Patos de Minas, Minas Gerais state, Brazil, evaluated the productivity, fruit maturation, and operation of mechanical harvesting of a coffee crop planted in four rows, with two faces of solar exposure and at two frequencies of rod vibration. Because of the regional conditions and culture, the planting alignments altered the levels of low-production harvests and the maturation of the fruit produced in high-production harvests, but no differences were observed between the faces of the plants. The increased frequency of the rod vibration varied according to the planting alignments in the low-production harvest, and increased harvesting capacity and the amount of picked mature fruits, reducing the losses of the remaining coffee of the high-production harvest.

Key words: *Coffea arabica* L., ripeness stage, sunlight, agricultural mechanization, coffee production, harvesting loses.

INTRODUCTION

In the coffee production system (*Coffea arabica* L.), the harvest and postharvest stages have several factors to be analyzed, because coffee is one of the few agricultural products that is priced based on qualitative criteria and whose value increases significantly with improved quality (Oliveira et al., 2007a). According to Pimenta and Vilela (2003), harvesting Brazilian Arabica coffee at different stages of maturation can affect the qualitative properties of this crop in the international market.

With the expansion of coffee growing in the Cerrado regions, the mechanization of agricultural operations is a key factor for Brazil to continue leading the world in

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coffee production (Oliveira et al., 2007c). Studies have shown that mechanized coffee harvesting represents 30% of the cost of production and 40% of the manpower employed, and increasing harvesting efficiency directly relates to cost reductions, without impairments and loss of the production system or damages to the crop (Santos et al., 2010).

The mechanical harvesting process of coffee is characterized by removal of the fruits from the plant by a procedure called strip-picking, which is highly complex and occurs through the use of vibration of rods (Aristizábal et al., 2003a). According to Barbosa et al. (2005), the vibration of the rods on the plants is an efficient method for harvesting coffee; however, the defoliation caused by this method forces the coffee tree to use reserves to restore vegetation, resulting in a lower subsequent production.

The level of solar radiation is an environmental variation that physiologically affects coffee plants and causes the plants to create adaptation mechanisms that can consequently interfere with fruit yield and maturation. Studies reported by Pinto et al. (2006) found a deleterious effect of light exposure to the afternoon sun with reduced growth and incidence of pests and diseases in a region with a high average temperature.

Researchers such as Pimenta and Vilela (2003), Silva et al. (2006), Oliveira et al. (2007a, b, c), Queiroz et al. (2007a, b) and Santos et al. (2010) have been studying the operational performance of cropping systems, and the locations and conditions of farming to reduce the cost of coffee production by reducing the time and number of harvests. However, there are few studies about the vibration frequency of the rods under conditions of sun exposure regarding the production and maturation of the fruits or about the rates of crop losses and the damages caused to the plants.

Thus, assuming that the sun exposure of the plants alters the maturation of the culture and that the frequency of the rod vibration could optimize indexes and reduce crop losses, this study evaluated the productivity, fruit maturation and operation of mechanical harvesting of the coffee crop in four planting rows, with two faces exposed to the sun and two rod vibration frequencies.

MATERIALS AND METHODS

This experiment was conducted at São João Grande Farm, located in Patos de Minas city, in the Savana's region of the Minas Gerais state, Brazil, during the harvest seasons of 2009/2010 and 2010/2011, in a circular planting area under a central pivot. This study used cultivar Catucaí Vermelho 785-15, which was six years old at the first evaluation, with a spacing of 4.00 m between rows and 0.50 m between plants, totaling 5,000 plants ha^{-1}. The geographic coordinates are approximately 18°33'18" south latitude and 46°20'01" west longitude, 6% slope, at an average elevation of 1,100 m, and the climate is rated as Cwa, according to the Köeppen method.

The tests were carried out with a mechanized harvesting of two crops. The 2009/2010 crop was named "low production" (negative biannuality) and 2010/2011 crop was named "high production" (positive biannuality). The harvest dates were defined based on the rate of green fruits in the plant. The first harvest was made on 06.04.2010, with an average of 14% green, 21% cherry and 65% coco grains in the plant; and the second was made on 05.21.2011, with an average of 10% green, 65% cherry and 25% coco. These plants were considered appropriate for harvesting, because the percentage of green fruits should be below 20% (Pimenta and Vilela, 2003).

Machinery

For the mechanized coffee harvest, a Jacto harvester, model KTR, was used. This harvester was made in 2003 and had approximately 4,000 h of functioning. This harvester operated displaced and attached to the hydraulic system of a three-point Massey Ferguson tractor, model 265 4x2TDA, with 47.8 kW (65 hp) of power in the motor to 36.6 Hz (2,200 rpm), and start-up is achieved through TDP at 9 Hz (540 rpm).

The harvester has a frame gantry and runs mounted ("over the row") over the plants among the coffee rows, with two picking cylinders equipped with vibrating rods that involve the coffee plant laterally, strip-picking the fruits through the effect of the vibration of the rods. The fruits fall into the collection system and are simultaneously discharged into a transfer set after cooling. During the harvesting operation, all the rods of the two cylinders of the harvester were used with the average speed fixed at 0.48 m s^{-1} (1.74 km h^{-1}), and the harvest was always performed in the same direction.

Statistical analysis

The experimental design was similar to a randomized block design. For crop productivity, four alignments (axes) of planting (A) were adopted, and the internal and external faces of the plants were separated from the center pivot (F). A factorial scheme of 4 × 2 with ten repetitions was analyzed, totaling 80 sample cells containing three plants in a row.

Since in earlier experiments no significant differences were detected regarding the production between the internal and external facing plants, the variables related to this mechanical harvesting factor were disregarded. Two frequencies of rod vibration (F) in alternating rows of harvest were tested, and four planting axes (A) in a 2 × 4 factorial arrangement with five replications were compared, which totaled 40 sample cells containing five plants in a row (Figure 1).

Four rows of planting (axes) inside the assessed area (A) were marked to identify the influence of sun exposure on the uniformity of fruit maturation and, consequently, the harvesting operation. Alignments were denominated as axes I to IV, with the positions varying by 45 degrees. The I axis is considered by Pinto et al. (2006) as the most favorable cultivation condition of the region, because the rays of the sun are over the tops of the plants for most of the day. In Axis III, the sun shines on only one side of the plant for part of the day and then shines on the opposite side for the rest of the day. Axes II and IV are considered intermediates to the aforementioned situations.

During mechanized harvesting, the vibration frequencies were 12.5 and 15.8 Hz (750 and 950 cycles/min). These frequencies were selected based on the vibration used at the moment of harvesting in the studied area (F1) and the results obtained by Oliveira et al. (2007b) (F2).

The crop productivity was initially determined by manual strip-picking of three plants in each cell sample, and the amount of coffee produced on each side of the plants was measured (L $plant^{-1}$). To determine the losses in the harvesting operation, the soil

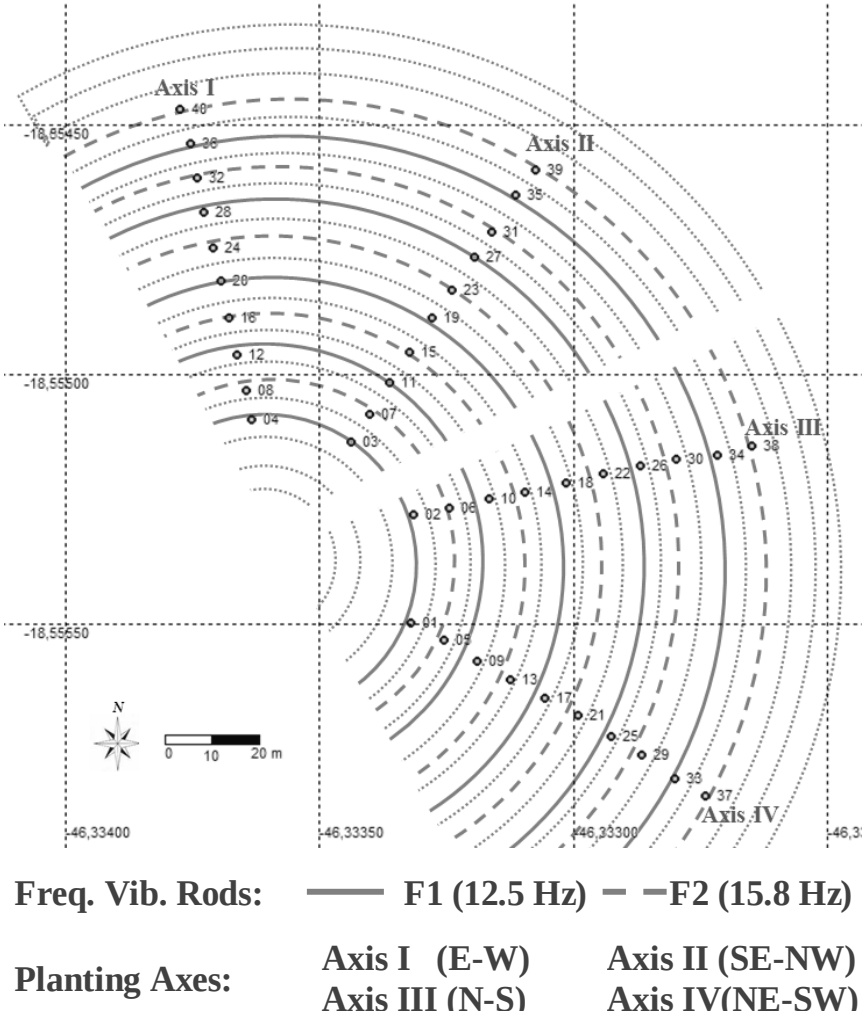


Figure 1. Georeferenced sampling grid, indicating the planting alignments and frequencies of the rod vibration.

beneath the top of five trees was lined with cloths of manual picking for each cell, and after the passage of the harvester, the volume of fallen fruit was measured (L plant^{-1}). Based on the productivity of the cell sample, the percentage of coffee dropped was determined.

The remaining volume of coffee was manually picked from the same five plants and was added to the volume of fallen fruit to determine the percentage of remaining coffee. The percentage of coffee harvested by the operation was determined by the difference between the yield and the loss rates obtained in each cell sample. Samples were taken from 1 L of coffee of all variables for the classification of fruit maturation stages (green, cherry and coco); the mature fruits, the portion of fruit at cherry and coco stages (Ch + Co), are of greater interest (Silva et al., 2006).

Defoliation caused by mechanical operation was quantified by the mass of leaves and fallen branches (g plant^{-1}) on picking cloths after the passage of the harvester and was determined when quantifying the loss of coffee.

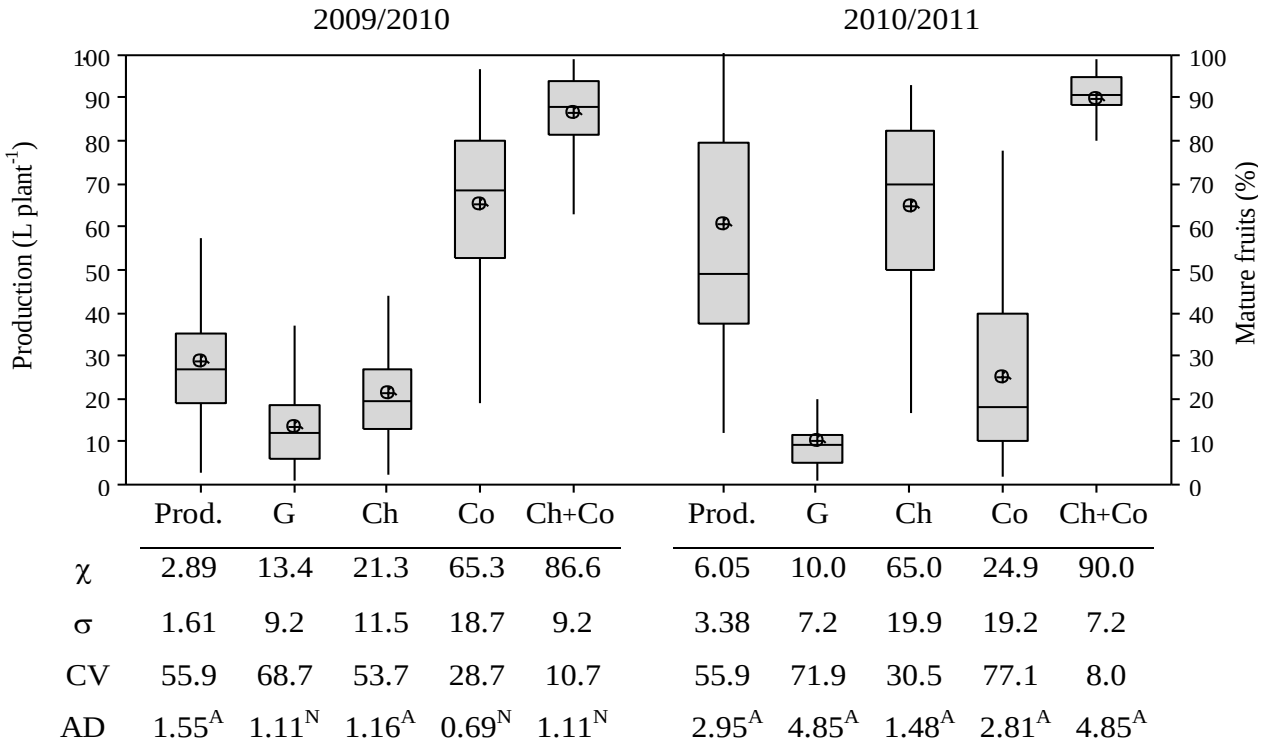
The results were statistically analyzed with the program Minitab® 16, which was passed through an exploratory analysis (descriptive statistics) to ensure normality of data, or the need for transformation to normalization. The variability behavior and the occurrence of disparate data are illustrated by boxplots.

The boxplots represent the distribution of a set of data based on the median (Q2), the lowest quartile (Q1), the higher quartile (Q3), interquartile range (IQR = Q3 - Q1) and minimum and maximum values of a parameter. This analysis illustrates the symmetry and the dispersion of the data, and it shows the presence or absence of disparate data. Boxplots are especially suitable for comparing two or more data sets corresponding to the categories of a variable.

A single factor analysis of variance (ANOVA) was conducted by applying the F test with a significance level of 5% to verify any significant differences between the averages of the variables. When appropriate, Tukey's test was applied at 5% probability to compare the means.

RESULTS AND DISCUSSION

The descriptive analysis (Figure 2) of the crop, showed an increase in the average of crop from 2009/2010 to 2010/2011. According to Oliveira et al. (2007a), this increase is due to culture biannuality, which demonstrates the stress caused by the production of a crop in the subsequent season. This phenomenon



	Prod.	G	Ch	Co	Ch+Co	Prod.	G	Ch	Co	Ch+Co
χ	2.89	13.4	21.3	65.3	86.6	6.05	10.0	65.0	24.9	90.0
σ	1.61	9.2	11.5	18.7	9.2	3.38	7.2	19.9	19.2	7.2
CV	55.9	68.7	53.7	28.7	10.7	55.9	71.9	30.5	77.1	8.0
AD	1.55^{A}	1.11^{N}	1.16^{A}	0.69^{N}	1.11^{N}	2.95^{A}	4.85^{A}	1.48^{A}	2.81^{A}	4.85^{A}

Figure 2. Descriptive statistics of the parameters related to coffee productivity and distribution of the maturation of fruits, in 2009/10 and 2010/11 crops. [1] Variables: Prod.: Production (L plant^{-1}); G: Green fruits (%); Ch: Cherry fruits (%); Co: Coco fruits (%); (C+P): Mature coffee, cherry fruits + coco fruits (%).[2]Parameters: χ: Arithmetic mean; δ: Standard deviation; CV: Coefficient of variation (%); AD: Anderson-Darling normality test. (N: normal distribution; A: asymmetrical distribution).

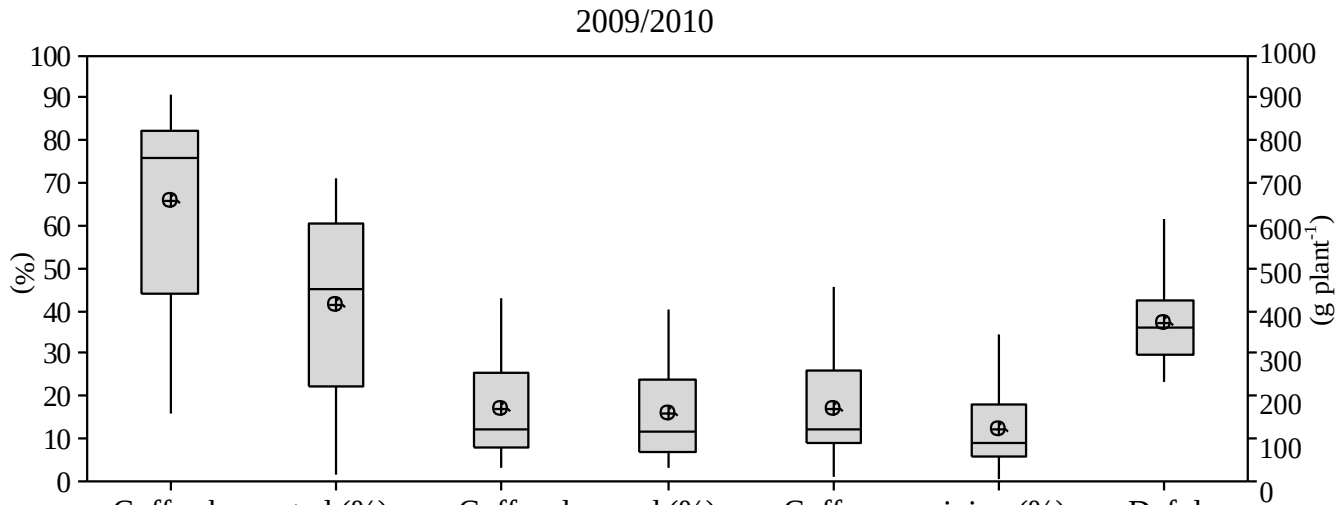
explained the repetition of the work in two seasons, and the high-production harvest obtained more than twice the average crop of low production harvest (2009/2010). Regarding the variability between samples, similar behaviors were observed between the two crops, with high coefficients of variation (Pimentel-Gomes and Garcia, 2002) and asymmetric distributions of the data by the Anderson-Darling test.

Regarding the distribution of fruit maturation, the 2009/2010 crop had a higher occurrence of coco fruits while the 2010/2011 crop had a higher occurrence of cherry fruits, which can give better qualitative properties (Queiroz et al., 2007a). Both harvests had a similar proportion of green fruits, which consequently resulted in the same proportion of harvested mature fruits (Ch + Co). Concerning the variability of the results, all stages of fruit maturation displayed differences between the averages and the median, with high values of standard deviation and coefficients of variation for both crop seasons. Only the results of green coffee and raisins in 2009/2010 had a normal distribution by the Anderson-Darling test. Only the sum of the mature fruits (Ch + Co) presented less variability, with low coefficients of variation and asymmetric distributions in the 2010/2011 crop.

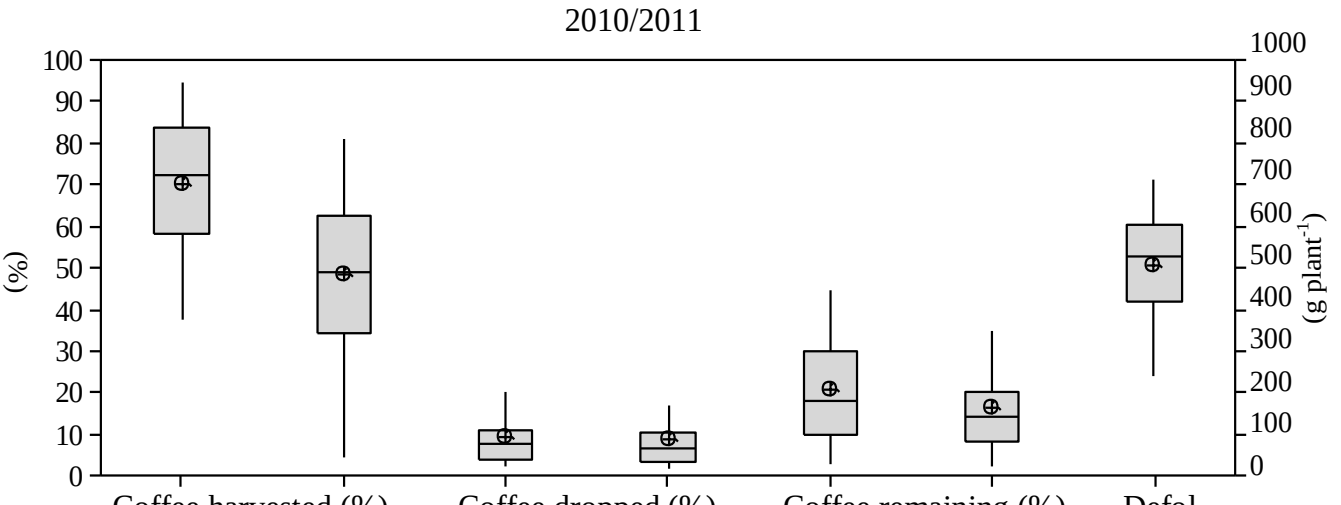
The descriptive statistics for the variables related to the mechanized harvesting of coffee in two crop seasons (Figure 3) indicate similar behavior for the two crops, with high amplitude data, high values of standard deviation and high coefficients of variation. The harvesting capacity of the operation might be considered insufficient (Oliveira et al., 2007b) for both seasons, because the averages were below 70%, which resulted in a loss of approximately 30%. These losses were distributed between coffee and plant remnants lying on the ground after a machine pass in the row.

Furthermore, the operation did not undertake a selective harvest, according to Silva et al. (2006). The production was, on average, 85% of mature coffee, while the coffee collection index was 60%, resulting in high rates of green fruits, which may influence the quality of the produced beverage (Queiroz et al., 2007a). Concerning the variability of the samples and the coffee collected, maturation also had high coefficients of variation, but still had normal distributions.

The losses of the fallen coffee and the remaining coffee were similar in the 2009/2010 crop, but the losses in the 2010/2011 crop were predominantly the remaining coffee in the plant. This result can be related to increased crop productivity and the greater availability of the fruits to be harvested (Silva et al., 2006). The coffee on the ground contained a higher concentration of mature fruit, and the fruit that remained on the tree contained a higher



	Coffee harvested (%)		Coffee dropped (%)		Coffee remaining (%)		Defol.
	Sum	(Ch+Co)	Sum	(Ch+Co)	Sum	(Ch+Co)	(g plant^{-1})
χ	65.96	41.53	17.28	16.10	16.76	11.99	374.7
σ	20.75	10.60	12.61	1.08	11.29	1.52	96.0
CV	31.45	27.45	72.93	6.78	67.38	12.67	25.62
AD	1.52^{A}	0.39^{N}	1.93^{A}	2.93^{A}	1.40^{A}	0.56^{N}	0.58^{N}



	Coffee harvested (%)		Coffee dropped (%)		Coffee remaining (%)		Defol.
	Sum	(Ch+Co)	Sum	(Ch+Co)	Sum	(Ch+Co)	(g plant^{-1})
χ	69.98	48.25	9.49	8.56	20.54	16.25	505.4
σ	17.47	11.44	9.40	0.41	14.19	2.85	114.9
CV	24.97	25.03	99.11	4.77	69.08	17.60	22.73
AD	0.47^{N}	0.99^{N}	3.58^{A}	1.02^{N}	1.12^{N}	1.49^{A}	0.59^{N}

Figure 3. Descriptive statistics of parameters related to coffee mechanized harvesting, fruit maturation and plant defoliation in 2009/2010 and 2010/2011 crops. [1]Parameters:χ: Arithmetic mean; δ: Standard deviation; CV: Coefficient of variation (%); AD: Anderson-Darling normality test. (N: normal distribution; A: asymmetrical distribution).

Table 1. Analysis of variance and average test for productivity and distribution of the coffee fruit maturation for 2009/2010 and 2010/2011 crops.

Factor	2009/2010					2010/2011				
	Prod.	G	Ch	Co	Ch+Co	Prod.	G	Ch	Co	Ch+Co
Faces (F)										
Internal	2.66	14.6	22.6	62.8	85.4	6.80	9.98	64.1	25.9	90.0
External	3.12	12.3	20.0	67.7	87.7	5.29	10.1	65.9	24.0	89.9
Axle (A)										
I	3.19	13.0	22.8	64.2	86.9	4.71	12.8^{a}	65.7	21.5	87.2^{b}
II	2.24	10.9	19.3	69.9	89.1	5.90	10.5^{ab}	66.8	22.7	89.5^{ab}
III	3.25	15.9	24.1	59.9	84.1	6.36	6.7b	64.7	28.6	93.3^{a}
IV	2.88	13.9	19.1	67.0	86.1	7.21	10.0^{ab}	62.9	27.0	90.0^{ab}
F test (P<0.05)										
F	1.67^{ns}	1.35^{ns}	1.08^{ns}	1.06^{ns}	1.11^{ns}	3.58^{ns}	0.09^{ns}	0.09^{ns}	0.37^{ns}	0.01^{ns}
A	1.68^{ns}	1.11^{ns}	1.04^{ns}	1.45^{ns}	1.35^{ns}	1.85^{ns}	3.38*	0.14^{ns}	0.60^{ns}	2.40*
F × A	0.77^{ns}	2.59^{ns}	1.30^{ns}	2.13^{ns}	2.59^{ns}	0.13^{ns}	0.89^{ns}	0.08^{ns}	0.51^{ns}	0.63^{ns}

[1]Variables: Prod.: Production (L plantc^{-1}); G: Green fruits (%); Ch: Cherry fruits (%); Co: Coco fruits (%); (Ch+Co): Mature coffee, cherry fruits + coco fruits (%).[2]In each column, for each factor, means followed by the same letters do not differ by the Tukey test at 5% probability. ^{ns}No significance; *Significant at 5% probability by the F test.

concentration of green fruit, because mature fruits are more easily detached from the plants than the green fruits (Ciro, 2001).

The variability of the loss results showed similar behavior in the two crops, with high standard deviations and coefficients of variation and an asymmetric Anderson-Darling distribution. The distribution of maturation of loss rates showed a variability ranging from low to medium and a normal distribution only for the coffee losses during the 2009/2010 and 2010/2011 crop, respectively.

Defoliation caused by the plant operation was higher in the high production harvest, which is a phenomenon of biannuality culture (Oliveira et al, 2007a). However, the mean values were inferior to the results obtained by Silva et al. (2006) and Oliveira et al. (2007b), which showed defoliation of approximately 800 g plant^{-1} in a similar model. Regarding the variability of the samples, although the values of standard deviation and coefficient of variation were high, the results showed normal distributions.

The analysis of variance and the average test for variables related to crop productivity and distribution of fruit maturation (Table 1) showed no difference between the planting alignments and the faces of the plants for the variables evaluated in the 2009/2010 crop. These results suggest that, despite the low productivity, there was uniformity in the volume of coffee produced and fruit maturation across all insulation conditions.

Similar behavior was observed in the 2010/2011 crop, even with a higher production volume (high-production year). This variable remained stable between the different planting alignments and for different faces. There was no difference between the evaluated axes for the ratio of green and mature coffee (Ch+Co) produced. The highest concentration of mature coffee was on Axis III in relation to the Axis I, which may be related to increased sunlight on the side of the plant during the day (Pinto et al., 2006).

The analysis of variance and the means test for the variables in relation to the mechanized coffee harvesting, fruit maturation and defoliation (Table 2) showed that, for the 2009/2010 crop, the rod vibration frequency showed interaction evaluated with the axes, and then the splitting was carried out (Table 3). The evaluated axes differed in the maturation of the harvested coffee, resulting in lower maturation for Axis I. The fallen coffee losses were greater for axes I than for axes II and III, although there was no difference between the axes to mature in this crop.

None of these factors significantly influenced the defoliation of the plants. The defoliation rates were low, even with the increased vibration frequency, which is directly related to the damage caused the harvester (Aristizábal et al., 2003b). This behavior can be explained by the reduced productivity of the crop, which according to Santos et al. (2010) stimulates the stems less.

The interaction deployment for the harvested coffee and the remaining coffee in the plant (Table 3) showed that the increase in the rod vibration frequency provided a smaller amount of coffee harvested in Axis IV, which according to Barbosa et al. (2005), may be related to crop losses. Between the rows of planting, a smaller amount was harvested from Axis I than axes II and III at the higher frequency, which is linked to the loss results of

Table 2. Analysis of variance and the average test for the parameters of the mechanized harvesting of coffee, fruit maturation and defoliation of plants, in 2009/2010 crop.

Factor	Coffee harvested		Coffee dropped		Coffee remaining		Defoliation
	Sum	(Ch+Co)	Sum	(Ch+Co)	Sum	(Ch+Co)	(g plant^{-1})
Rod vibration frequency (F)							
12.5 Hz	68.0	42.4	13.8	12.5	18.2	13.3	349.4
15.8 Hz	63.9	35.1	20.8	19.4	15.3	10.7	400.0
Axle (A)							
I	52.6	24.4^{b}	27.6^{b}	25.8	19.8	14.0	405.7
II	67.2	41.5^{a}	14.2^{a}	12.9	18.6	13.8	370.1
III	74.6	47.4^{a}	12.3^{a}	11.2	13.1	9.3	347.6
IV	69.4	43.4^{a}	15.1^{ab}	14.0	15.5	10.9	375.4
F test (P<0.05)							
F	0.69^{ns}	2.86^{ns}	4.14^{ns}	1.73^{ns}	0.94^{ns}	1.29^{ns}	2.64^{ns}
A	3.16*	3.21*	3.04*	0.32^{ns}	0.67^{ns}	0.45^{ns}	0.59^{ns}
F × A	3.21*	1.43^{ns}	1.58^{ns}	1.18^{ns}	5.27*	2.23^{ns}	0.24^{ns}

[1]In each column, for each factor, means followed by the same letters do not differ by the Tukey test at 5% probability. ^{ns}No significance; *Significant at 5% probability by the F test.

Table 3. Deployment of the interaction between the factors for the harvested and remaining coffee in the plant, in the 2009/2010 crop.

Coffee harvested (%)	Vibration frequencies (F)	
Axle (A)	**12.5 Hz**	**15.8 Hz**
I	61.4^{Aa}	43.8^{Ab}
II	59.3^{Aa}	75.1^{Aa}
III	68.7^{Aa}	80.5^{Aa}
IV	82.6^{Aa}	56.3^{Bab}
Coffee remaining (%)	**Vibration frequencies (F)**	
Axle (A)	**12.5 Hz**	**15.8 Hz**
I	17.3A^{ab}	22.4^{Aa}
II	29.0^{Aa}	8.1^{Ba}
III	17.0^{Aab}	9.3^{Aa}
IV	9.5^{Ab}	21.5^{Aa}

[1]For each variable, the means are followed by the same capital letters with row, and lowercase letters in the column show a non-significant result by Tukey's test at a 5% probability.

the fallen coffee described in Table 2.

On Axis II, the higher rod vibration frequency increased the picking ability of the harvester. The amount of coffee remaining on Axis IV was lower than that on Axis II for the lowest vibration frequency.

Analysis of variance in the 2010/2011 crop (Table 4) showed that in years of high crop production, although there were differences in fruit maturation in productivity (Table 1), the planting alignments did not influence the variables in relation to the crop, the distribution of mature plants or the defoliation of the plants. These results suggest a uniformity in the operation, regardless of the insulation conditions of the culture.

The increase in the rod vibration frequency raised the rates and maturity of the harvested coffee, thereby reducing the losses by the remaining coffee on the plants after the passage of the mechanized harvester. This result demonstrates the increased capacity of picking of the harvester by increasing the vibration frequency without increasing damage to the crop, which has been

Table 4. Analysis of variance and the average test for the parameters of the mechanized harvesting of coffee, fruit maturation and defoliation of plants, in 2010/2011 crop.

Factor	Coffee harvested		Coffee dropped		Coffee remaining		Defoliation
	Sum	(Ch+Co)	Sum	(Ch+Co)	Sum	(Ch+Co)	Sum
Rod vibration frequency (F)							
12.5 Hz	64.3[b]	37.6[b]	10.9	9.9	24.7[a]	19.8	483.7
15.8 Hz	75.6[a]	54.6[a]	8.0	7.1	16.3[b]	12.6	527.1
Axle (A)							
I	70.6	45.5	8.9	7.8	20.5	15.9	564.5
II	75.5	53.2	7.7	6.9	16.8	12.7	508.1
III	63.3	38.8	9.8	9.1	26.9	21.8	503.3
IV	70.5	45.8	11.5	10.4	17.9	14.5	445.7
F test (P<0.05)							
F	4.38*	7.97*	0.68[ns]	1.55[ns]	4.32*	0.35[ns]	1.43[ns]
E	0.87[ns]	0.63[ns]	0.17[ns]	2.19[ns]	1.27[ns]	0.31[ns]	1.78[ns]
F × E	0.64[ns]	0.64[ns]	0.75[ns]	0.95[ns]	2.62[ns]	0.15[ns]	0.05[ns]

[1]In each column, for each factor, the means are followed by the same letters were not significantly different according to Tukey's test at a 5% probability; [ns]No significance; *Significant at 5% probability by the F test.

directly linked to the action of vibration of the rods (Oliveira et al., 2007a), and this observation agrees with the results of previous research (Ciro, 2001; Aristizábal et al., 2003a; Barbosa et al., 2005; Silva et al., 2006; Oliveira et al., 2007b; Santos et al., 2010).

Conclusion

The harvesting operation proved inadequate for the two crops analyzed, with a low rate of harvested mature fruit. Planting alignments altered the rates of the harvest in the crop of low production and maturation of fruits produced in high-production harvest, with no differences between the rows facing different directions.

The effects of the increased rod vibration frequency varied according to the planting alignments in the low-production harvest and increased the harvesting capacity by increasing the amount of picked mature fruit and by reducing the losses of the remaining coffee in the high-production harvest.

Conflict of Interests

The authors have not declared any conflict of interests.

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Full Length Research Paper

Germination and emergence of *Mouriri elliptica* mart., a rare medicinal fruit tree native to the Brazilian Cerrado biome

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This study is aimed to describe the germination process of *Mouriri elliptica* Martius, a species in the Melastomataceae family that is native to the *Cerrado* (tropical savannah) and popularly known in Portuguese as *croada*, *coroa de frade* [friar's crown], *croadinha* [little croada], *puçá-preto* [black net], or *jabuticaba-do-Cerrado*. The species has significant potential for use as food and medicine; however, there is a dearth of scientific studies on this plant. Two experimental tests were performed to clarify the germination process of this species: Germination was evaluated *in vitro* by integrating fruit ripening and culture medium, and the emergence and initial growth of *M. elliptica* Mart. seedlings obtained from seeds soaked for different lengths of time were observed in a greenhouse. It was verified that the *in vitro* environment was not effective for germination independent of the culture medium, even using known and effective methodologies for breaking dormancy in seeds of this species. For the emergence of seedlings, it was found that soaking seeds in distilled water for two days was the optimal length of time for seedling production.

Key words: Breaking dormancy, Melastomataceae, propagation, tetrazolium test.

INTRODUCTION

Mouriri elliptica Mart. is popularly known in Portuguese as *coroa de frade* [friar's crown] or *croadinha*, and in a study by Rufino et al. (2011), the species *M. elliptica* Mart. is referred to as *puça* [net]. This plant belongs to the family Melastomataceae; it can reach 4 to 6 m in length and is found in the Brazilian states of Goiás, Mato Grosso and Mato Grosso do Sul (Silva et al., 2001). According Silva et al. (2001), this plant has occurence in Cerrado and

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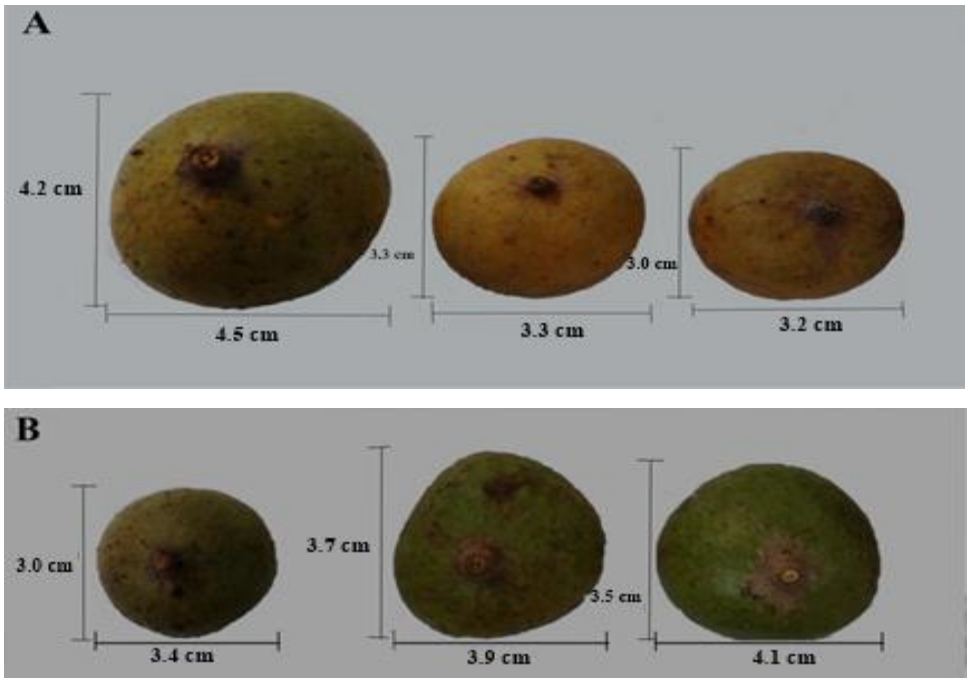

Figure 1. *Mouriri elliptica* Mart. fruits. (A) Ripe Fruits with partially orange epicarp coloration; and (B) unripe fruits with partially green epicarp coloration.

cerradão fitofisiony, yout frutification occur at September and December. According to the International Union for Conservation of Nature (IUCN, 2014), certain species of the genus are considered threatened with extinction, including *Mouriri completens* (Pitt.), *M. laxiflora* (Morley), *M. gleasoniana* (Standl.) and *M. panamensis* (Morley); however, *M. elliptica* Mart. does not appear to be threatened at this time.

The food and medicinal potential of *M. elliptica* Mart. Has been empirically established and was first described by Almeida et al. (1998) and Silva et al. (2001), who highlighted the lack of research. Currently, the shortage of publications on this plant could reflect the difficulty of finding it in its natural habitat. Articles in relevant journals are restricted to phytosociological surveys of the Pantanal biome and characterizations of its potential for vitamin C production and as an ulcer therapy (Moleiro et al., 2009; Rufino et al., 2011).

Interest in the conservation of native Cerrado species prompted the study by Vasconcelos et al. (2010) on the dormancy and breaking of dormancy in *M. elliptica* Mart. seeds, which was the first such report and verified that soaking seeds in distilled water for up to 48 h and mechanical scarification increased germination rates compared with seeds that did not receive such treatment. However, the combined effect of soaking and scarification was not evaluated.

For the genus *Mouriri*, only any studies have been conducted on its germination and chemical composition (Salis and Mattos, 2010; Vasconcelos et al., 2010; Rufino et al., 2011; Silveira et al., 2013), and there is a lack of research related to *in vitro* propagation in this genus (comment author). Therefore, the aim of this study was to evaluate the influence of the fruit ripening stage as well as the influence of seed scarification and soaking on the germination of seeds *in vitro* and in a greenhouse.

MATERIALS AND METHODS

Tests were performed in the Plant Tissue Culture Laboratory of the Federal Institute of Education, Science and Technology of Goiás, Rio Verde Campus, Goiás (GO), Brazil (*Instituto Federal de Educação, Ciência e Tecnologia Goiano, Câmpus Rio Verde*, GO). The *M. elliptica* Mart. fruits for Test 1 were collected in October 2013, and fruits for Test 2 were collected in November of the same year in the district of Planalto Verde, municipality of Montividiu, GO, Brazil at 17º 19.201' S, 51 33.500' W and 982 m.a.s.l.

A digital caliper was used to obtain biometric parameters, which included the mass (g) and longitudinal and equatorial diameter of the fruits and seeds. The ratio between the equatorial diameter and longitudinal diameter was obtained to determine the fruit shape.

Test 1: *In vitro* germination of *Mouriri elliptica* Mart. seeds in different concentrations of sucrose and fruit ripening

M. elliptica Mart. seeds were obtained from ripe fruits (epicarp partially orange) (Figure 1A) and unripe fruits (epicarp partially green) (Figure 1B). After biometric analyses of the fruits, they were manually de-pulped using a sieve and running water.

Once de-pulped, four hundred seeds were used, being two hundred from mature fruits and two hundred green fruits, were washed in running water and subsequently subjected to a

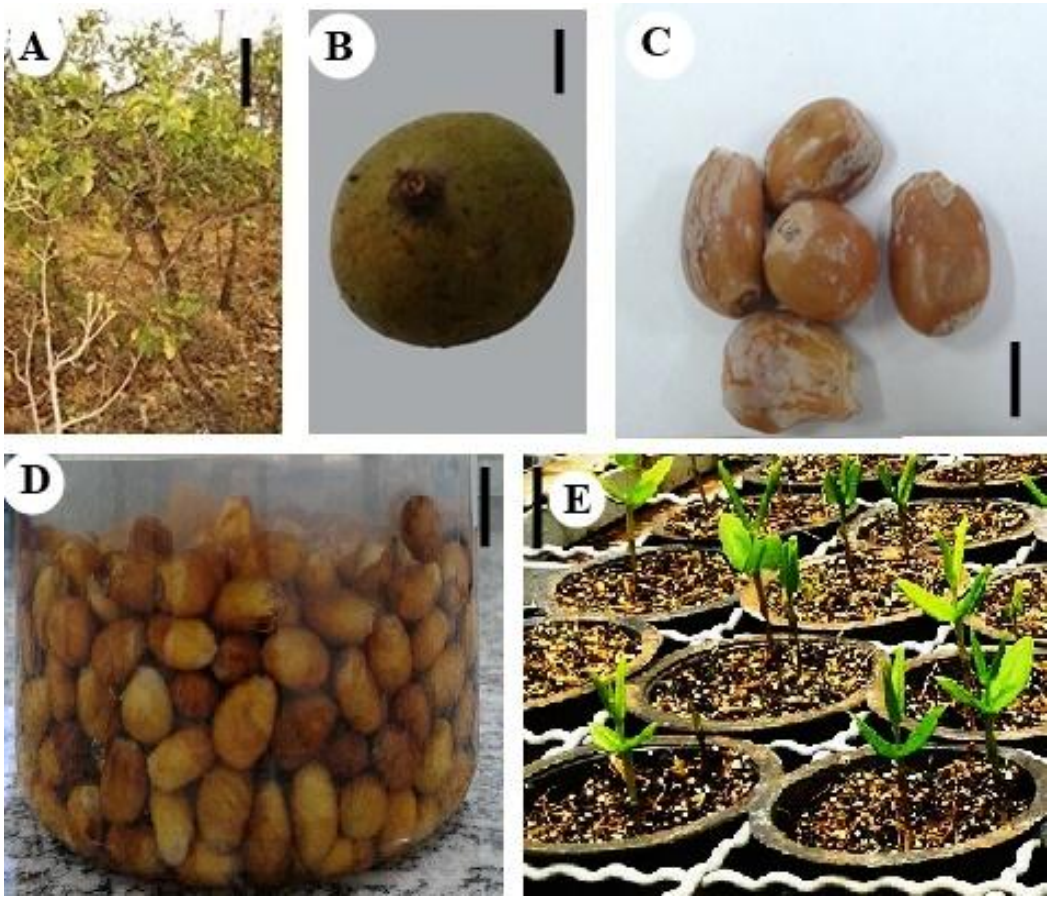

Figure 2. (A) Collection the mother plant; bar = 0.5 m. (B) Ripe fruit; bar = 11.2 mm. (C) Scarification of seeds with #60 sandpaper; bar = 10.0 mm. (D) Soaking of seeds for up to six days in distilled water at 30°C; bar = 25.0 mm. (E) Emergence of *Mouriri elliptica* Mart. seedlings in the greenhouse; bar = 2.8 cm.

dormancy breaking test following the methodology of Vasconcelos et al. (2010). Briefly, the seeds were soaked in distilled water for 48 h at room temperature (25±3°C); disinfected in a laminar flow cabinet, with the seeds remaining for one minute in 70% alcohol and four minutes agitated in commercial 100% sodium hypochlorite (Qboa, Industries Anhembi, Curitiba-PR, Brasil); and then rinsed three times with distilled and autoclaved water. For *in vitro* germination, the seeds were inoculated in test tubes containing 10 mL distilled and autoclaved water in 0, 30, 60 and 90 gL^{-1} sucrose concentrations. To avoid submersion of the seeds, "germitest" paper was used as a support. The seeds remained in these conditions for 60 days in a grow room at a temperature of 25 ±3ºC and photoperiod of 16 h.

The percent germination and seed contamination were evaluated at 60 days after inoculation (DAI). The experiment was conducted in an completely randomized design with a 2 × 4 factorial design of maturation [2] and sucrose concentrations [4], with four repetitions. To investigate seed viability at the end of the test, a tetrazolium test with four repetitions of ten seeds was performed. A tetrazolium solution of 0.075% was used at 30ºC for 24 h.

Test (2): Scarification and soaking of *M. elliptica* Mart. seeds

Seeds from ripe fruits (Figure 2B) and de-pulped fruit (with the aid of a sieve) were used. To evaluate the effect of scarification (Figure 2C), one group of seeds was sanded opposite the hilum using 60 grit sandpaper based on the techniques of Santos et al. (2004). To evaluate the effect of soaking, the seeds were maintained in a beaker containing distilled and autoclaved water and placed in a Mangelsdorf germinator at a temperature of 30ºC forzero, two, four and six days, with water changed every two days (Figure 2D).

To determine the soaking curve, four repetitions of ten seeds were used. The seeds were weighed hourly for the first six hours, every two hours for the next six hours and every 12 h until reaching 72 h from the beginning of the soaking process.Subsequently, the seeds were weighed daily. To determine the water content, the greenhouse method 105±2°C was used, and the wet weight was used as a base until a constant mass was achieved.

After the established soaking times had elapsed, four samples from 15 seeds were placed in plastic tubes containing the commercial substrate Trimix® andmaintained in a greenhouse with three daily irrigations. Before seeding, the seeds were treated with fungicide [active ingredients(carboxin + thiram): 200+200 gL^{-1}] at doses of 500 mL to 100 kg seeds and 500 mL distilled water to 100 kg seeds (Fungicide, Chemtura Indústria Química, São Paulo – SP, Brasil).

An evaluation was performed 60 days after seeding (DAS), and the percent emergence, length and numbers of leaves were analyzed (Figure 2E). The experimental design was entirely

Table 1. Mass of unripe and ripe fruits from Test 1 and ripe fruits and seeds of Test 2 of *Mouriri elliptica* Mart. Equatorial diameter andlongitudinal diameter. Rio Verde, Goiás, 2014.

	Test 1 - October collection			
	Equatorial diameter (mm)	**Longitudinal diameter (mm)**	**Mass (g)**	**Ratio (e.d./l.d.)**
Ripe fruits	35.5 ± 0.67 ^{ns}[1]	28.27 ± 0.50^{ns}	21.96±1.19^{ns}	1.25
Unripe fruits	34.94 ± 0.60	29.1 ± 0.42	21.43±0.95	1.20
Test 2 - November collection				
Ripe fruits	34.89 ± 0.63	28.67 ± 0.41	20.5±1.01	1.22
Seeds	11.9 ± 0.16	15.77 ± 0.21	1.14±0.04	0.75

[1]±Standard error of the mean. ^{ns}Not significant by t test with 5% de probability.

Table 2. Percent viability of *Mouriri elliptica* Mart. seeds from Test 1 after the tetrazolium test.

	Vigor	**Viable (%)**	**Unviable (%)**	**Dead (%)**
Unripe	0	5	0	95 ± 2.89[1]
Ripe	0	10	0	90 ± 4.08

[1]±Standard error of the mean.

randomized in a factorial arrangement of two(scarification) by four (soaking times of zero, two, four and six days), with four repetitions of 15 seeds.

RESULTS AND DISCUSSION

Biometrics

The biometric analyses showed diameters with homogenous values. In Test 1 (Table 1), the fruits had an average equatorial diameter of 35.22 mm, longitudinal diameter of 28.68 mm and mass of 21.69 g. In Test 2, homogeneity of the fruit diameters was also verified. According to the biometric parameters, *M. elliptica* Mart. fruits have an oval shape and are flattened, which was confirmed by the ratio of diameters that reached values above 1.20.

Test 1: *In vitro* germination of *Mouriri elliptica* Mart. in different concentrations of sucrose and fruit maturation

After 60 days of cultivation, *in vitro* germination of the seeds was not observed; therefore, it was not possible to fit a mathematical model to explain the data as a function of increases in sucrose concentration, due to low germination percentages. However, a high percentage of contamination (30.62%) was observed.

The high rate of contamination observed in this study could be the result of the disinfection method and high resistance and impermeability of the tegument. To guarantee more effective disinfection, Couto et al. (2004) removed the tegument of seeds, which resulted in minimal contamination of 9.52% during the *in vitro* germination of seeds of mahogany (*Swietenia macrophylla* King).

The resistance of the tegument implies a lack of water absorption, which is necessary in the germination phase. Because no studies have reported on the *in vitro* micropropagation of this species, additional tests should be developed to investigate the efficacy of breaking tegumentary dormancy and achieving *in vitro* germination.

Because *in vitro* germination was not demonstrated, the tetrazolium test was performed (Table 2), which identified seed viability at 10% from ripe fruits and only 5% for seeds from unripe fruits. Thus, performing the tetrazolium test according to the adapted methodology permitted sufficient coloration to distinguish vigor; however, the seeds suffered from high mortality in this cultivation environment (Figure 3).

Test 2: Scarification and soaking of *M. elliptica* Mart. seeds

According to an analysis of variance, differences did not occur in mass and water content between scarified and intact seeds. An increase in these variables was observed in the first hour, with mass values of 11.04 to 13.07 g for scarified seeds and 8.28 to 9.7 g for non-scarified seeds (Figure 4A) and water content values (Figure 4B) from 19.01 to 31.61% in scarified seeds and 17.52 to 29.54% in non-scarified seeds. After the first hour of soaking, the mass values and water content remained stable.

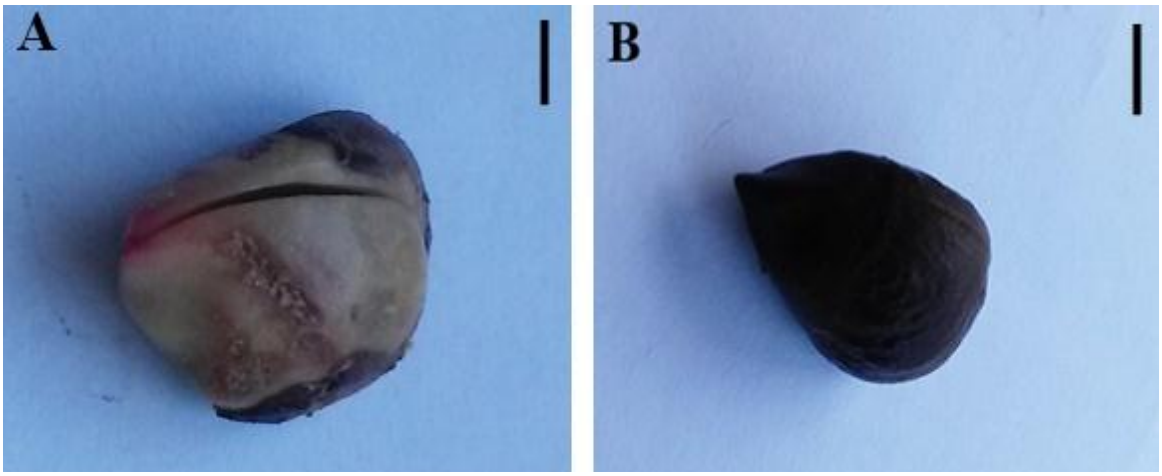

Figure 3. Seeds of *Mouriri elliptica* Mart.after the tetrazolium test. (A)Viable seeds; bar = 4.5 mm. (B) Dead seeds; bar = 4.1 mm. Rio Verde, Goiás, 2013.

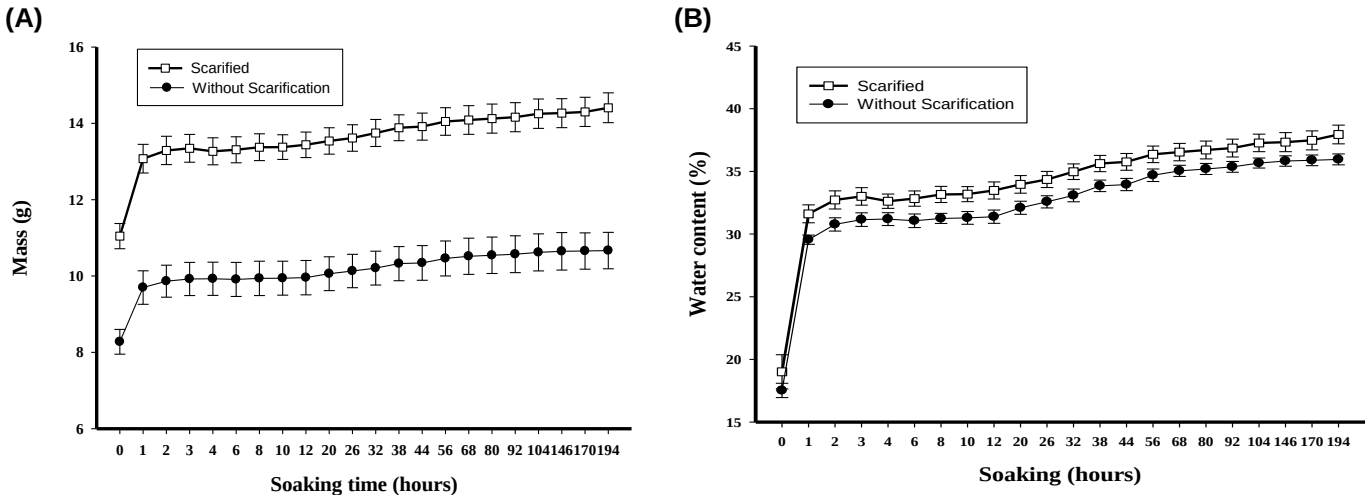

Figure 4. Soaking of scarified and intact *M. elliptica* Mart. seeds. A) Mass gain of seeds (g). B) Water content of seeds (%).

There was an interaction between soaking time and scarification that influences the emergence and length of seedlings. However, only soaking time affected the number of seeds.

The scarified seeds could not be fit to a mathematical model to explain the behavior of the data on seedling emergence and length, which reached an average of 50.83% and 4.12 cm, respectively (Figure 5A to B). For non-scarified seeds, an increase in percent emergencewas observed after soaking the seeds for two to six days, whereas a greater length (4.6 cm) was observed in the seedlings obtained from seeds soaked for two days. A decrease in the number of leaves was observed with an increase in soaking time, with values of 2.90 cm at time zero and 1.76 cm at six days (Figure 5C).

For *M. elliptica* Mart., gibberellic acid, sulfuric acid and water immersion, cooling and heating and mechanical scarification were studied by Vasconcelos et al. (2010), who achieved success with pre-soaking in gibberellic acid, soaking in water for 48 h and mechanical scarification of seeds. However, increased soaking times, which is a simple and inexpensive method, were not evaluated; thus, the study presented here is essential, and the results verified that an increase in soaking time of non-scarified seeds for up to six days has an impact on seedling emergence in this species, which exhibits tegumentary dormancy.

The efficacy of breaking tegumentary dormancy by mechanical scarification was effective for the species *Stryphnodendron adstringens* Mart.,*Dimorphandra mollis* Benth,*Sterculia foetida* L. and *Bowdichia virgilioides* Kunth., showing that this type of dormancy is common in

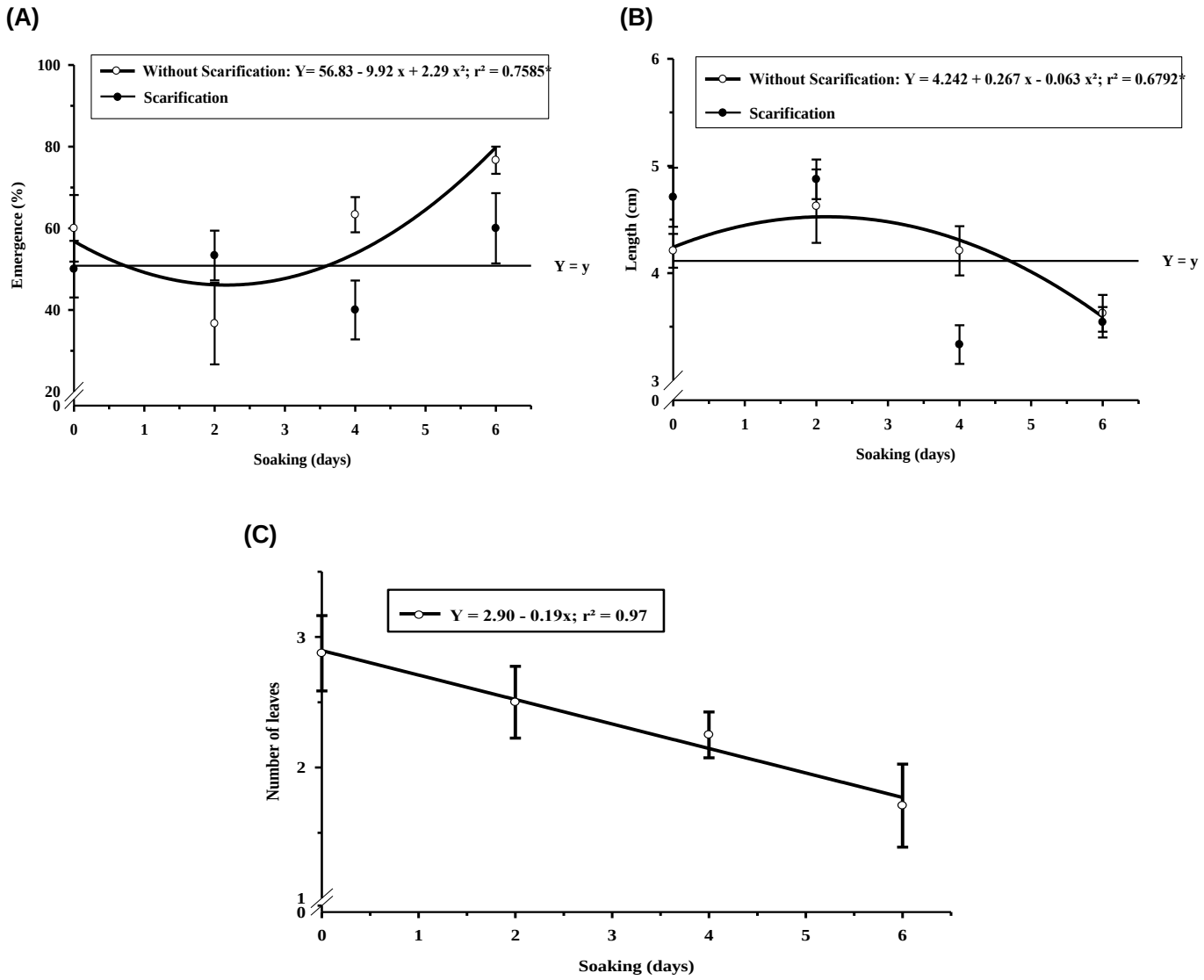

Figure 5. Effect of soaking scarified and intact seeds on the (A) emergence, (B) length of seedlings and(C) number of leaves per seedling of *M. elliptica* Mart.

native species of the Cerrado (Santos et al., 2004; Albuquerque et al., 2007; Martins and Nakagawak, 2008; Oliveira et al., 2008).

Conclusion

In vitro germination of *M. elliptica* Mart. seeds did not occur, and this behavior was independent of fruit ripeness and sucrose concentration in the culture medium. In non-scarified *M. elliptica* Mart. seeds, increased values for percent emergence occurred with increases in seed soaking time for up to six days. Seedlings with a greater leaf number and length were obtained from scarified seeds and seeds soaked for zero and two days, respectively.

Conflict of Interests

The authors have not declared any conflict of interests.

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Escherichia coli O 157 in curd cheese

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The microbiological quality of curd cheese prepared in northeastern Brazil and sold in street markets and supermarkets in the towns of Feira de Santana and Cruz das Almas BA Brazil, was evaluated for mesophyllic and psycrotrophic count, molds and yeasts, total coliforms and *E. coli*, and the occurrence of *E. coli* O157. Sixty-eight curd cheese samples were collected between February and June 2014 and the analysis for each microorganism was based on Norm N. 62 published in August 26, 2003 by the Ministry of Agriculture, Livestock and Supply (MAPA), according to successive dilutions, time and temperature of each organism. Counting of colonies was expressed as colony-forming units per ml of sample (UFC / mL). *Escherichia coli* O157 were transferred to the Singlepath *E. coli* O157 test from the culture medium MEC Broth with Novobiocin. The curd cheese revealed 100% contamination by mesophyllic microorganisms, 51.47% by psycrotrophic, 98.53% by yeasts and molds, 88.24% by coliforms, 77.94% by *E. coli* and 22% by *E. coli* O157 serotype. Curd cheese sold in the street market and in supermarkets in Feira de Santana and Cruz das Almas proved to have been prepared under unsatisfactory hygienic conditions, endangering the health of consumers and featuring contamination rates above those permitted by law.

Key words: *Escherichia coli*, hygiene, bacteria, fungi, coliforms, microorganisms.

INTRODUCTION

Foodborne diseases are still one of the major public health problems world wide and account for considerably high cases of illness. Recent reports indicate that *Staphylococcus aureus*, *Salmonella* spp., *Listeria monocytogenes* and pathogenic *Escherichia coli* are considered the most frequent pathogens and they are responsible for outbreaks mainly associated with raw milk and raw or inadequately heated milk products (Ahmed and Shimamoto, 2014; Feitosa et al., 2003; Fernandes et al., 2006; Oliveira et al., 2014).

The main reservoir of this pathogen appears to be wild and domestic ruminants such as deer, cattle, goats and sheep. In addition to undercooked beef hamburgers and other meat products, as well as water, fruits and vegetables, cheeses made from raw milk have been implicated in infections of *E. coli* O157:H7 (Brasil, 2003; Mora et al., 2007; Moreno et al., 2002; Oliveira et al., 2012; Öksüz et al., 2004). Since dairy cattle are asymptomatic carriers of E. coli O157:H7, meat and milk products are thought to be risky foods from the point

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of view of this organism. Therefore, insufficient heat-treatment of ground meat and raw milk forms a potential infection risk. The processing conditions for different milk products are very important from the standpoint of the organisms infection risk. Although, it can be destroyed with pasteurisation, the survival rate in other processes is high, for example it can tolerate 8.5% salt concentration. Cheese made with unpasteurised milk is potential vehicle for transmission of *E. coli* O157 to the consume (Oliveira et al., 2012, 2014; Otero et al., 2014).

Despite the risk presented by *E. coli O157:H7* in dairy products, there are few studies on the incidence of this microorganism in Brazil. The current analysis evaluates the microbiological quality of curd cheese commercialized in street markets and in supermarkets of Feira de Santana and Cruz das Almas BA Brazil, by counts of mesophyll and psycrotrophic microorganisms, molds and yeasts, total coliforms, *E. coli* and *E. coli* O157.

MATERIALS AND METHODS

Sixty-eight curd cheese samples were collected from the street market and supermarkets of Feira de Santana and Cruz das Almas, in the northeastern state of Bahia, Brazil, between February and June 2014. The samples were retrieved in aseptic conditions and transported in an isothermal container with recycled ice and maintained under refrigeration till laboratory analyses at the Animal Microbiology and Parasitology of the Universidade Federal do Recôncavo da Bahia UFRB, Cruz das Almas BA Brazil.

Microbiological analyses were conducted following recommendations and requirements by RDC n. 12 of 2/1/2001 (Blanco et al., 2003; Brasil, 2001a). The methodology for microbiological analyses was foregrounded on Norm 62 of 26/8/2003 of the Brazilian Ministry of Agriculture, Livestock and Supply (MAPA) which provided the official analytic methods for the microbiological analyses for the control of animal-derived products and water (Brasil, 2001a, b, c).

The microbiological analyses performed were: counts of mesophyll and psycrotrophic microorganisms, filamentous fungi (molds) and yeasts, total coliforms, *E. coli* and confirmation of *E. coli* O157.

Plate count agar (PCA) culture was employed to count viable strict and facultative aerobic mesophyll and psycrotrophic microorganism. Cultures were incubated in a culture buffer and fridge according to the time and temperature of each microorganism. Filamentous fungi (molds) and yeasts counts were undertaken with Dextrose Sabourand Agar culture medium incubated in a BOD-like buffer. The colonies for each microorganism were counted and multiplied by the corresponding dilution factor and the result given as colony-forming units per mL of the sample (CFU/mL) (Ahmed and Shimamoto, 2014; Apha, 1998).

Total coliform and *E. coli* were counted in Chromocult Coliform Agar culture medium and incubated in a culture buffer. The simultaneous detection of total coliforms and *E. coli* was undertaken with a combination of two types of chromogenic substrates. The salmon substrate β-D-GAL was cleaved by the enzyme β-D-galactosidase, characteristic of other coliforms, resulting in colonies with salmon to red coloration. The X-β-D-glucuronide substrate was cleaved by β-D-glucuronidase enzyme, characteristic of *E. coli*, resulting in colonies with dark blue to violet coloration (ISO 7218:1996 / 1:2001). *E. coli* O157 was identified by the immunochromatographic fast test to detect *E. coli* O157 (SINGLEPATH *E. COLI* O157 EMB. C/25 TESTES MERCK) (Ahmed and Shimamoto, 2014; Apha, 1998).

Brain heart infusion (BHI) broth was used for the propagation and maintenance of *E. coli* colonies which, after inoculation and incubation in a buffer, were placed on Petri plates with Nutrient Agar culture medium for conservation and maintenance. The cultures were maintained under refrigeration for 5 months till collection was concluded. Colonies from the nutrient agar were then inoculated in MEC broth with Novobiocine (MERCK) and incubated (Ahmed and Shimamoto, 2014). They were later transferred to Singlepath *E. coli* O157 test, a fast immunochromatic test for the detection of *E. coli* O157, with a circular sampling point, with the oval form as test (T) and control (C) zones. When a drop from the MEC broth culture with Novobiocine is placed, the assay works correctly if a red line appears on the control zone (C) within 20 min. The sample is thus positive for *E. coli* O157 if prior to 20 min the red lines appear in the two zones (T and C). The sample is negative when no red line appears on the test zone and it fails to appear clearly on the control zone (C) during 20 min after the application of the sample.

Microbiological data were transformed into log UFC/g. Minimum rate, maximum rate, arithmetic average, variance, coefficient of variation were performed by absolute and percentage distribution for nominal variables and statistical measures. Tukey's test verified the significant difference in the detection of microorganisms at the different collection sites.

Contamination rates on the street market and in supermarkets of Feria de Santana and Cruz das Almas were similarly measured by the analysis of variance for mesophyll and psycrotrophic microorganisms, fungus molds and yeasts, total coliforms and E. coli. Although means did not differ statistically ($p<0.01$), or rather, at 1% significance, Tukey´s test was applied to compare averages between the treatments. Absolute rates of the samples exhibited the same letter.

Table 1 demonstrates that the differences in the coefficient of variation (CV) occurred for total coliforms among the samples from the two towns. Feria de Santana (FSA – SM) had CV equal to 35.32%, followed by 16.89% for FSA - S, whereas in Cruz das Almas, CV were 25.34% and 11.15% respectively for CA – SM and CA - S. Therefore, curd cheese in the supermarkets of Cruz das Almas (CA - S) had more homogeneous rates as compared to samples from other collection places under analysis. However, standard deviation (SD) of samples from the supermarkets of Cruz das Almas (CA - S) had the lowest rate (0.60) and thus revealed that differences were not so big.

Although in the case of E. coli, rates on the street market in Feira de Santana (FSA - SM) had a lower CV (21.90%), the samples of the street market of Cruz das Almas had the lowest standard deviation (SD) or rather, 0.66. The above revealed that its samples were more homogeneous when compared with those from other sampling sites.

RESULTS AND DISCUSSION

S. aureus and *E. coli* in cheese are frequently used as indicators of hygienic quality and show lack of microbiological safety. Some strains of these organisms are known to cause diseases in humans. Therefore, they are undesirable in high number (Otero et al., 2014; Öksüz et al., 2004; Salotti et al., 2006).

The microbiological evaluations of curd cheese revealed microorganism counts which indicated total coliforms and *E. coli*; deteriorating microorganisms such as mesophylls and psycrotrophic microorganisms, molds and yeasts; and pathogenic microorganisms such as

Table 1. Results of standard deviation and coefficient of variation of total coliforms and *E. coli* analyzed in the towns of Feria de Santana and Cruz das Almas.

Microorganisms (log UFC/g)	Standard deviation (S)	Coefficient of variation (CV) (%)
Total Coliforms (FSA – SM)	2.07	35.32
Total Coliforms (FSA – S)	1.28	16.89
Total Coliforms (CA – SM)	1.30	25.34
Total Coliforms (CA – M)	0.60	11.15
E. coli (FSA – SM)	0.96	21.90
E.coli (FSA – S)	1.18	46.64
E.coli (CA – SM)	0.66	27.73
E. coli (CA – S)	1.50	51.37

FSA – Feira de Santana; FL – street market; S – Supermarket; CA – Cruz das Almas.

E. coli O157, given in Figures 1 - 5 and Tables 2 and 3.

All the microorganisms under analysis showed 100% contamination by mesophyll microorganisms; 51.47% by psycrotrophic microorganisms; 98.53% by molds and yeasts; 88.24% by total coliforms; 77.94% by *E. coli*; 22% by serum type *E. coli* O157. The intake of curd cheese with the above improper conditions may have serious consequences in the population and thus, public health is involved. According to RDC n. 12, the minimum limit is the acceptable lot of the product, whilst the maximum limit separates the acceptable from the non-acceptable product. Intermediate rates between the maximum and minimum limits are products of intermediate acceptable rates making mandatory the hygiene conditions in the processing of the products (Blanco et al., 2003; Brasil, 2001a).

The curd cheese analyzed in the two towns had mesophyll bacteria counts between 4.32 and 13.05 log CFU/g (Figure 1). Curd cheese samples were exposed on the trays of the street markets and in supermarkets, without any proper hygiene, causing the contamination.

Count rates of mesophyll bacteria in the current experiment revealed only two samples with 4.32 and 4.97 log FCU/g and thus within the limits recommended by ANVISA, following RDC n. 12 of 2/1/2001, which accepts mesophyll microorganisms up to 5 log FCU/g (Blanco et al., 2003; Brasil, 2001a, b, c).

High mesophyll rates in food indicate that they were prepared with highly contaminated raw matter, that processing was unsatisfactorily from sanitary aspects and that food was stored in inadequate time and temperature conditions. However, high levels of mesophyll bacteria were not used to analyze the sanitary conditions of fermented lactic products in which microorganisms are used in the processing (Kousta et al., 2010).

When compared with the mesophyll microorganism rates, the psycrotrophic microorganisms had lower rates, or rather, between 1 and 4.62 log CFU/g (Figure 2). Only one sample among the samples analyzed for Cruz das Almas was contaminated with 1 log CFU/g, whereas Feira de Santana had 55.7% of samples with the bacterium. Brazilian legislation fails to delimit psycrotrophic microorganisms in milk products. Microbiological contamination caused by processing and/or storage flaws may have contaminated the product, coupled with its organoleptic characteristics (taste, aroma, color and texture) (Apha, 1998).

With the exception of AMT 17, molds and yeasts were detected in curd cheese (Figure 3). The other samples were above the tolerance limit of 2 log CFU/g recommended by RDC n. 12 of 2/1/2001. According to Lourenço and Sousa (2001) analyses on samples of curd cheese from Marajó, Brazil, produced from buffalo and cow milk showed a rate >3 log UFC/g for the same microorganism.

Samples of curd cheese in Feira de Santana showed between 4.23 and 11.88 log CFU/g, whereas in Cruz das Almas the rates ranged between 8.27 and 9.41 log CFU/g. Mold and yeast rates in curd cheese revealed contamination in the product and thus health risks to the consumer, due to higher rates than those established by sanitary legislation. Re-contamination of curd cheese may occur during handling in the final processing and in storage. Since some molds are psycrotrophic, their spores survive and germinate in favorable conditions. Refrigeration temperature inhibits mold development but they are incapable of destroying them. Anaerobiosis may improve the product´s conservation (Loguercio et al., 2001).

Figures 4 and 5 show log CFU/g rates of total coliforms and *E. coli*, with 11.76 and 22% of the 68 samples under analysis within the tolerance margin; the other samples revealed <1 log CFU/g of total coliforms (Brasil, 2001). According to Salotti et al. (2006), the human intestine and that of other animals is the habitat of this bacterium group. Incorrect handling and faulty application of procedures of good manufacture practice are involved when it occurs in food. It is thus indicative of fecal contamination with health risks to consumers due to its association with pathogenic microorganisms. Kousta et al. (2010) underscore that the presence of these microorganisms in cheese evidences pasteurization flaws

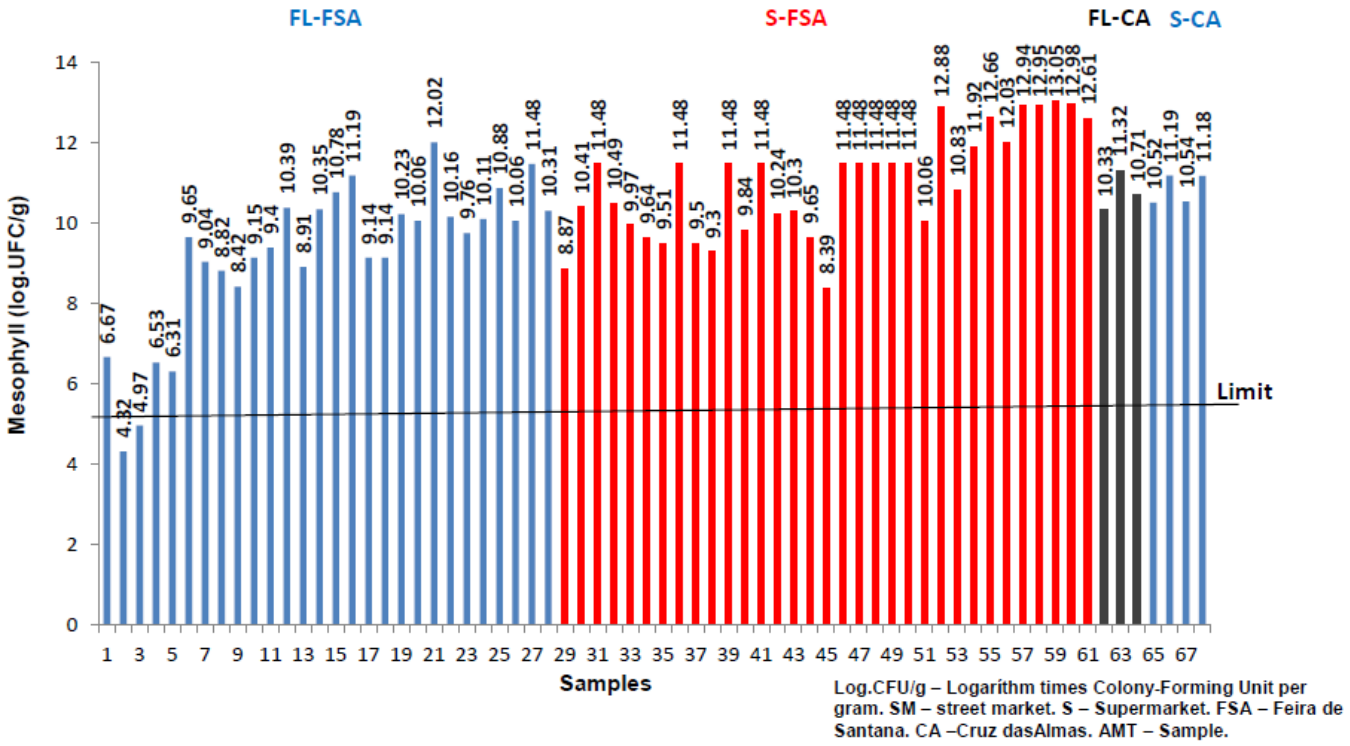

Figure 1. Concentration rates of mesophyll microorganisms from curd cheese produced in Feira de Santana and Cruz das Almas BA Brazil, February – June, 2014.

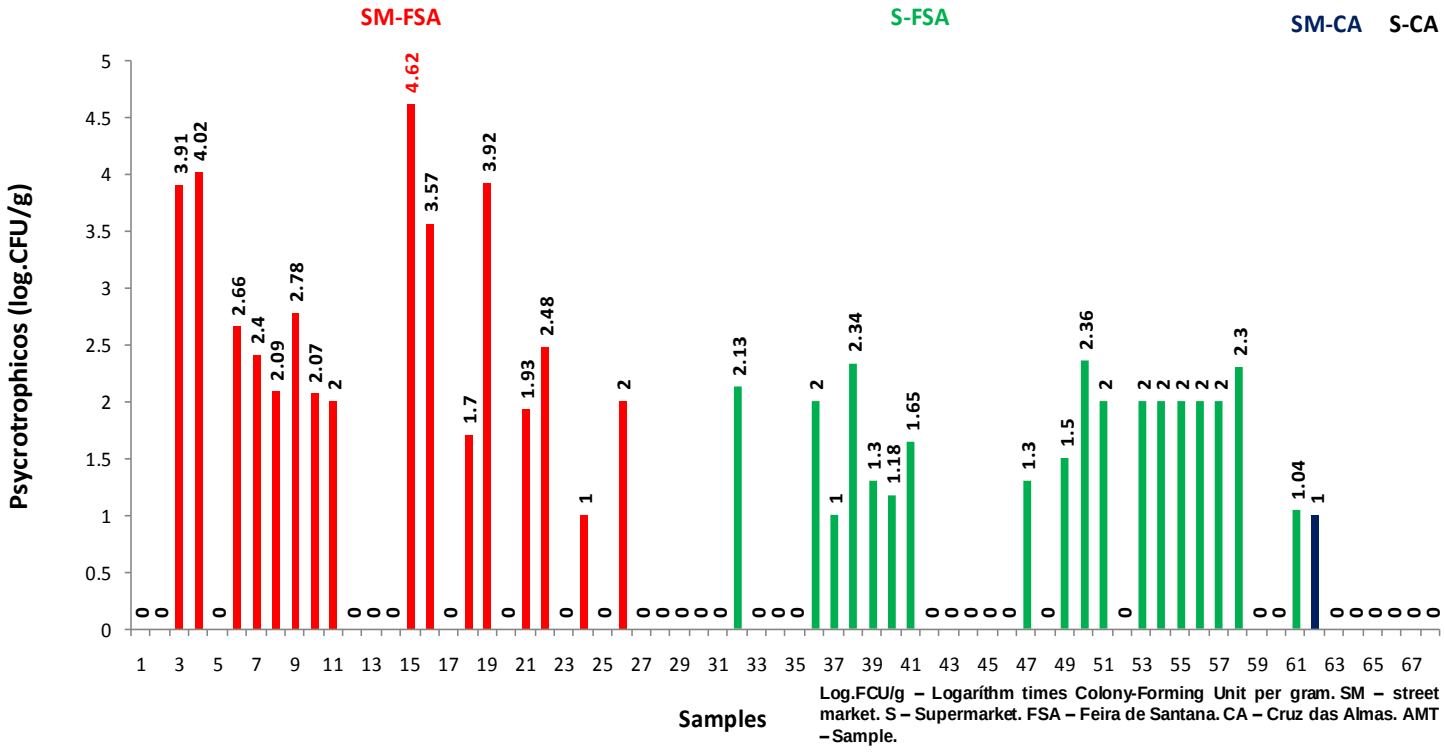

Figure 2. Concentration rates of psycrotrophic microorganisms from curd cheese samples produced in Feira de Santana and Cruz das Almas BA Brazil, February – June, 2014.

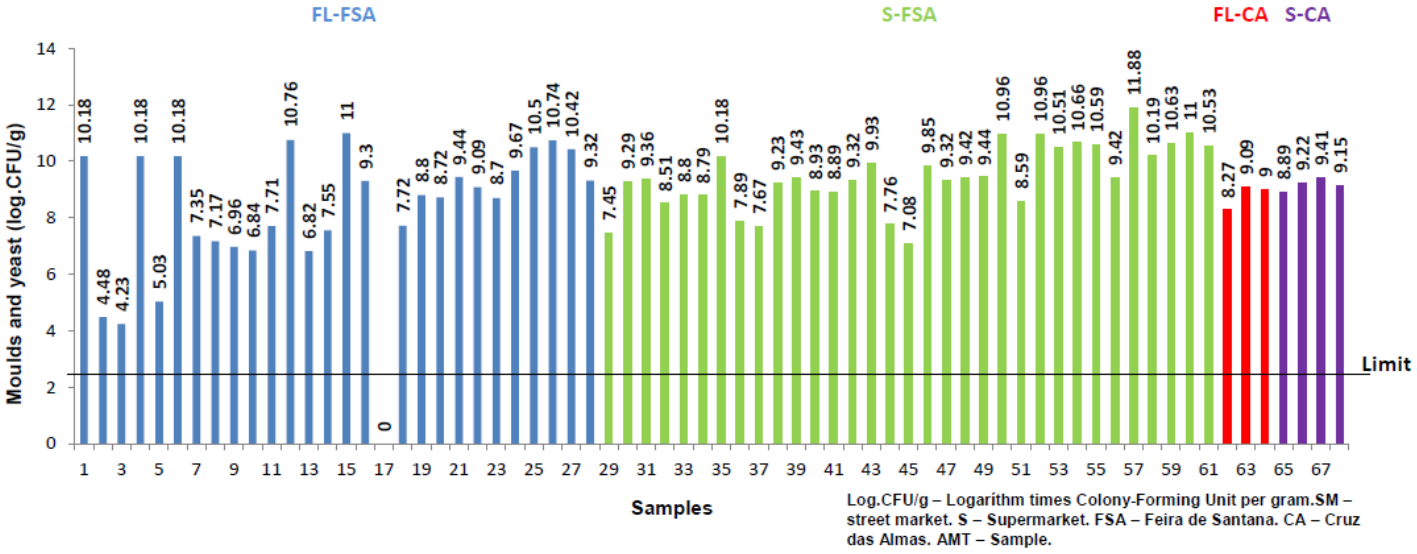

Figure 3. Concentration rates of molds and yeasts in samples of curd cheese produced in Feira de Santana and Cruz das Almas BA Brazil, February – June, 2014.

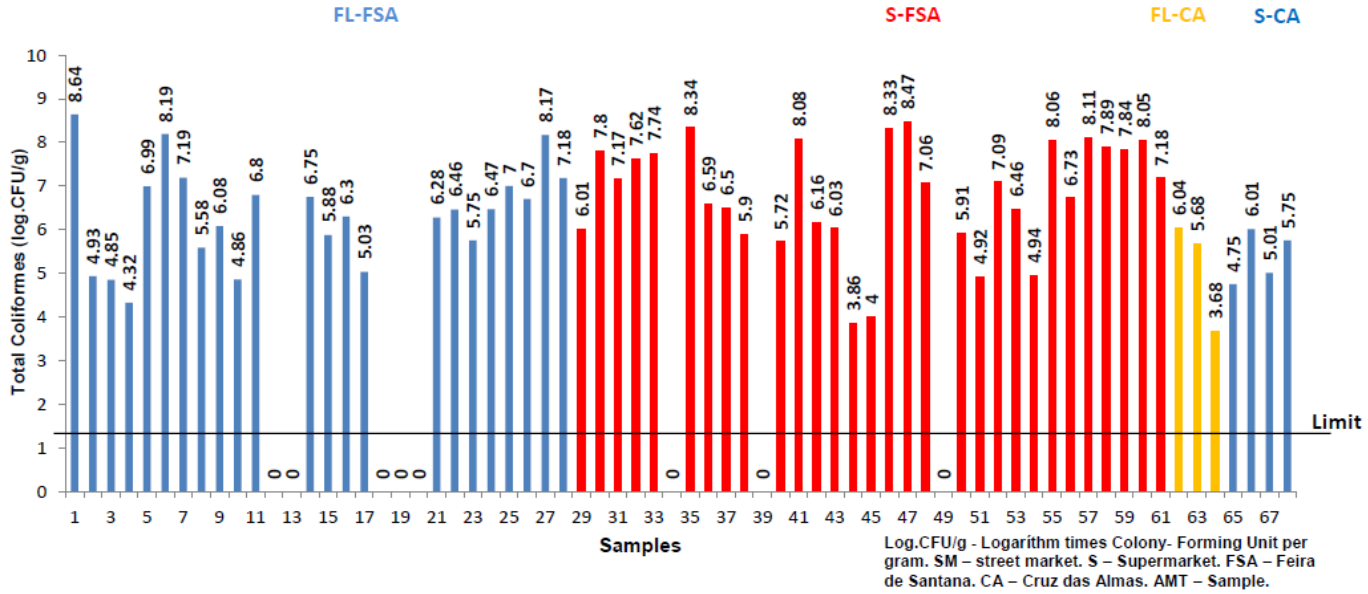

Figure 4. Concentration rates of total coliform bacteria in samples of curd cheese in Feira de Santana and Cruz das Almas BA Brazil, February – June, 2014.

or post-pasteurization contamination due to the fact that correct pasteurization eliminates all indicator and pathogen microorganisms.

The presence of 100% total coliforms, with counts ranging between 3.68 and 6.04 log UFC/g, occurred for curd cheese in Cruz das Almas. *E. coli* was not merely detected in one sample; since the others were above the limit established by Brazilian legislation, unsatisfactory sanitary hygiene conditions were rampant.

Analyses showed that most curd cheese comer-cialized failed to have the required conditions for consumption. High contamination index by microorganisms in the curd cheese may have occurred due to faulty prime matter up to an adequate place for selling the product. In fact, all the cheese samples were exposed without any protection or refrigeration, with the consequent survival and multiplication of microorganisms (Mora et al., 2007). Contamination by air is an important issue in product

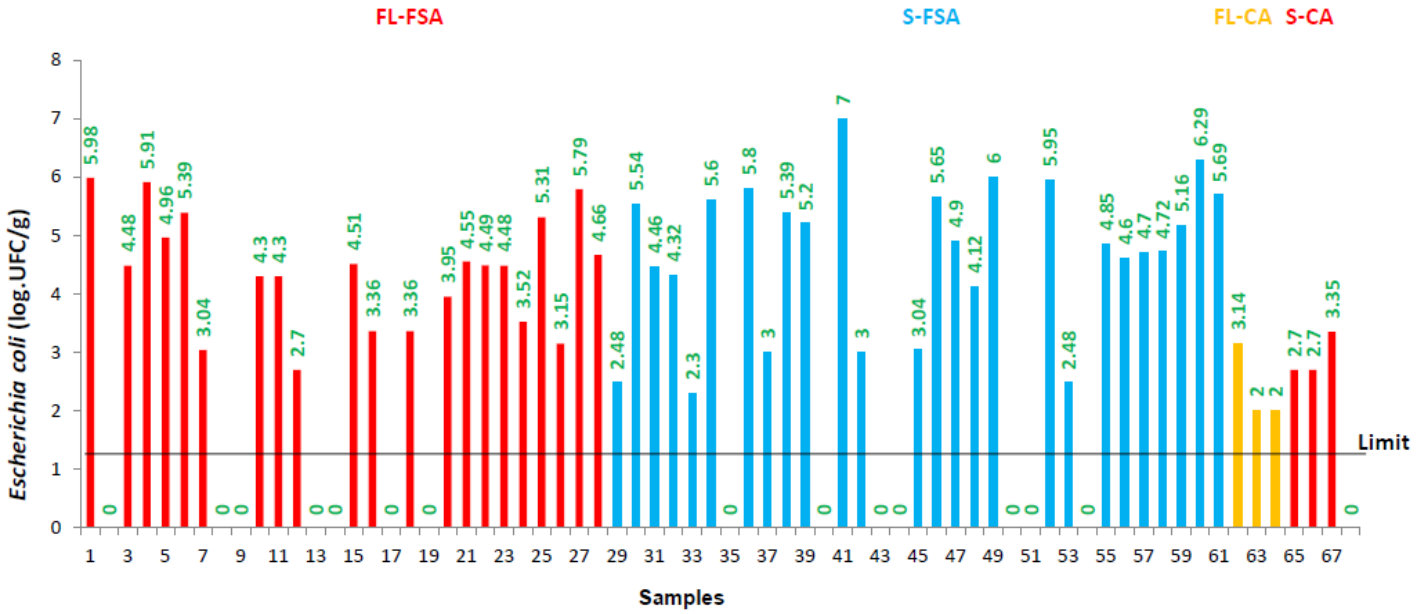

Figure 5. Concentration rates of bacteria *Escherichia coli* from curd cheese samples in Feira deSantana and Cruz das Almas BA Brazil, February – June, 2014.

Table 2. Presence or absence of *Escherichia coli* O157 in samples of curd cheese in Cruz da Almas BA Brazil. February - June 2014.

Samples	Place	Presence/absence
AMT 62	SM	-
AMT 63	SM	+
AMT 64	SM	+
AMT 65	S	-
AMT 66	S	+
AMT 67	S	-
AMT 68	S	

SM– street market; S– supermarket.

production units whose concern lies in quality control of their products. The air is a vector without any specific flora but has a great amount and variety of suspended microorganisms, or rather, they are maintained in the air by dust and liquid particles (Fernandes et al., 2006).

Cheeses commercialized on street markets and in supermarkets of the two towns manifest unsatisfactory hygiene conditions, with great health risks for the consumer. The effective application of hygiene and sanitization principles recommended for the elaboration of cheese should be undertaken so that consumers may have products featuring acceptable microbiological quality, with a long shelf life.

Varying levels of presumptive *E. coli* counts in the samples may indicate the extreme diversity of hygienic practices undertaken throughout the milk and cheese productions. The incidence and presumptive *E. coli* counts are similar to those reported by Kaan and Özdemir (2006), generally made from raw milk, and reveals an extensive deficiency of satisfactory cheese manufacturing and/or post production handling, because these organisms are killed by pasteurization and are found in a wide variety of habitats sanitary practices during milk production (Öksüz et al., 2004; Salotti et al., 2006).

Tables 2 and 3 demonstrate the occurrence of *E. coli* O157 in curd cheese. *E. coli* is a pathogenic bacterium with serious repercussions in public health. Fifteen out of the 53 samples contaminated by *E. coli* were positive for *E. coli* O157. The percentage is very high since all food should be free from pathogenic microorganisms.

In many developed countries, O157:H7 is the most prevalent STEC serotype associated with severe disease in humans. However, estimation of the prevalence of O157-associated infections in Latin America is difficult since screening for O157 is performed by only a few microbiological laboratories from few countries (Loguercio et al., 2001).

Curd cheese is rich in nutrients and deteriorates fast by microorganisms if not stored properly. Hygiene from the production of prime matter to production, storage, transport and commercialization is basic. Contamination of curd cheese may occur in all these stages. Further, *E. coli* in food may be assessed under two headings. *E. coli* may be initially an enterobacterium and indicates fecal microbial contamination and consequently the product was prepared in unsatisfactory hygiene conditions.

Another aspect may be taken into account, that is, *E. coli*

Table 3. Presence and absence of *Escherichia coli* O157 in samples of curd cheese in Feira de Santana BA Brazil, February – June, 2014.

Samples	Place	Presence/absence	Samples	Place	Presence/absence
AMT 1	SM	-	AMT 29	S	-
AMT 2	SM		AMT 30	S	-
AMT 3	SM	+	AMT 31	S	-
AMT 4	SM	+	AMT 32	S	+
AMT 5	SM	-	AMT 33	S	+
AMT 6	SM	-	AMT 34	S	+
AMT 7	SM	-	AMT 35	S	
AMT 8	SM		AMT 36	S	-
AMT 9	SM		AMT 37	S	-
AMT 10	SM	-	AMT 38	S	-
AMT 11	SM	-	AMT 39	S	-
AMT 12	SM	+	AMT 40	S	
AMT 13	SM		AMT 41	S	+
AMT 14	SM		AMT 42	S	-
AMT 15	SM	+	AMT 43	S	
AMT 16	SM	-	AMT 44	S	
AMT 17	SM		AMT 45	S	+
AMT 18	SM	-	AMT 46	S	-
AMT 19	SM		AMT 47	S	-
AMT 20	SM	-	AMT 48	S	-
AMT 21	SM	-	AMT 49	S	-
AMT 22	SM	-	AMT 50	S	
AMT 23	SM	-	AMT 51	S	
AMT 24	SM	-	AMT 52	S	-
AMT 25	SM	-	AMT 53	S	-
AMT 26	SM	-	AMT 54	S	
AMT 27	SM	-	AMT 55	S	-
AMT 28	SM	+	AMT 56	S	-
-	-		AMT 57	S	-
-	-		AMT 58	S	-
-	-		AMT 59	S	+
-	-		AMT 60	S	+
-	-		AMT 61	S	-

SM– street market; S- Supermarket.

strains are pathogenic for humans and animals (Ahmed and Shimamoto, 2014; Leite Júnior et al., 2000; Loguercio et al., 2001).

The Single path *E. coli* O157 test, a fast immuno-chromatic test for the detection of *E. coli* O157, revealed that 15 samples (22%) showed the characteristic presence of the strain (Tables 2 and 3). Seven and eight samples of curd cheese sold on the street market and in the supermarkets of the two towns contained respectively, *E. coli* O157. This fact is of great concern since EGEC is an important microorganism which causes gastroenteritis in people at the extremes of the age bracket and in immunosuppressed patients (Fernandes et al., 2006). Enterohemorrhagic *E. coli* (EHEC) is pathogenic for humans and belongs to the shiga toxigenic *E. coli* (STEC) group (Ahmed and Shimamoto, 2014). It is one of the great causes of gastroenteritis, causing hemorrhagic colitis (HC) or hemolitic uremic syndrome (HUS), the main cause of kidney disease in children. Since its identification as a pathogen in 1982, STEC O157 has been held to be the cause of several diseases, especially in Canada, Japan, UK and US (Aquarone et al., 2001; Bergamini et al., 2007).

Food of animal origin has been identified as the main vehicle for the transmission of foodborne pathogens to humans. Three major foodborne pathogens affecting people worldwide are *S. enterica*, *E. coli* O157:H7 and *Shigella* spp. An overview of foodborne disease reports

from seven countries has indicated that milk and milk products were implicated in between and 5% of the total number of bacterial outbreaks. Several large outbreaks of foodborne disease due to the consumption of raw milk cheese have been reported. *E. coli* O157:H7 can potentially enter the human food chain from a number of animal sources. The presence of these pathogens can be due to contamination taking place during milking at dairy farms or because of poor handling of meat, milk and cheese b*y* retailers (Ahmed and Shimamoto, 2014; Otero et al., 2014; Öksüz et al., 2004).

Curd milk prepared in the northeastern region of Brazil is a very soft cheese, but also an excellent culture medium. Besides its high moisture index, it is rich in nutrients that enhance the growth of microorganisms. Curb cheese is thus easily contaminated by micro-organisms throughout its production process till commercialization. Refrigeration and vacuum packaging may be a strategy to reduce contamination since they delay the deterioration process of the curd cheese exposed during commercialization.

Results show that the samples analyzed for the micro-organisms researched in the current assay do not comply with Brazilian legislation and may be considered as low quality products. The curd cheeses analyzed are improper for intake since contamination causes risk to human health. It is safer to opt for inspected and refrigerated products, wrapped in proper packages, with the date of manufacture, best before date and conditioning.

In Brazil, domestically-produced raw milk cheeses are popular with consumers, so their availability is considerably higher than cheeses made from pasteurized milk. Consequently, educating employees, retailers and consumers on the appropriate handling and storage of meat and dairy products is essential to effectively prevent contamination. Simple intervention strategies, such as promoting hand washing with soap and good hygienic practices at the slaughterhouses, processing plants and retailer shops, can have a sound practical impact on public health. Health education for the general population is required by regulatory authorities where the sale of raw milk and rawmilk products is permitted. A strong science-based approach that addresses all the issues involved in continuing to improve food safety and public health is necessary to prevent foodborne illnesses. The strengthening of food control systems including foodborne disease surveillance and food monitoring is required, especially in developing countries for the prevention, detection and control of foodborne illnesses.

Conflict of Interests

The authors have not declared any conflict of interests.

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Full Length Research Paper

Biomass yield, nutrient content and *in vitro d*ry matter digestibility of morphological fractions of two sweet potato varieties intercropped with coffee or maize-haricot bean

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Effects of stages of growth and intercropping of two sweet potato varieties (Hawassi-83 and Tula) (alone or with coffee or maize-haricot bean) on biomass yield and *in vitro* dry matter digestibility (IVDMD) of morphological fractions were assessed in Southern Ethiopia. Similar coffee plantations (age, density, soil type, topography) were selected for intercropping. Vines were planted on three plots per intercropping type at 15 days interval, and cultivated three times. Morphological fractions were collected from 1.0 m^2 of land on 4th, 5th and 6th months for nutrient and partial budget analyses (revenue estimated for 100 m^2 of land). Ten elders evaluated varieties at 2nd, 4th and 6th months after planting. Lowest DM (77 g/kg) was obtained from fresh Tula stem-petiole (FTSP) but highest (190 g/kg) from fresh Tula leaf (FTL) ($p<0.05$). Fresh Tula vine (FTV) had lowest (840 g/kg DM) organic matter and FTL ($p<0.05$) highest (914 g/kg DM). Crude protein content of fresh Hawassi-83 stem-petiole (FHSP) was lowest (113 g/kg DM) but that of FTL highest (269 g/kg DM). Lowest vine yield (0.27 kg/m^2) was from Tula intercropped with coffee but highest (0.82 kg/m^2) from maize-haricot bean intercropping followed by sole Hawassi-83 (0.56 kg/M^2). Overall mean vine DM yield (333.6 g/m^2) of maize-haricot bean intercropping was greater ($p<0.05$) than that of intercropping (174.0 g/m^2) with coffee. IVDMD of fresh Hawassi-83 leaf (FHL) (78.0±0.27) and FTSP (77.0±0.10) were lowest but that of FHSP (84.3±0.59) and FTL (84.3±0.04) highest. Both Hawassi-83 and Tula vine were best produced when intercropped with maize-haricot bean, although Hawassi-83 was more profitable and preferred by respondents than Tula.

Key words: Intercropping, morphological fractions, nutrients, stage of maturity, sweet potato vine.

INTRODUCTION

Ethiopia owns 55,027,280 cattle, 27,347,933 sheep, 28,163,332 goat, 1,963,010 horses, 6,953,077 donkeys, 356,087 mules, 1,098,312 camels, 51,350,738 poultry and 5,052,297 beehives (CSA 2013/14) which play crucial role by contributing 15 to 17% of GDP, 35 to 49% of agricultural GDP and 37 to 87% of household incomes

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(ILRI, 2010).

The huge livestock population is constrained by insufficient and poor quality feed. Natural pastures and poor quality crop residues are the main sources of livestock feed in East Africa. However, grazing lands do not fulfill the nutritional requirement of animals particularly in dry seasons (Zinash et al*.,* 1995). With the rapid increase in human population and high demand for food, pastures are steadily being converted to crop lands. Less productive marginal lands unsuitable for cultivation are left for grazing. Deforestation and overgrazing has also substantially reduced soil fertility and productivity (Alemayehu, 1999). The limited land available for feed and fodder production and poor quality pasture demanded total dependence of livestock on crop residues and by-products (Firew, 2001). Assessing forage production options in the limited land available is of paramount importance to mitigate this constraint.

In this study, the effect of stage of growth and intercropping of sweet potato with coffee or maize-haricot bean on biomass yield of morphological fractions of sweet potato and profitability of intercropping were assessed.

MATERIALS AND METHODS

Description of the study area

The study was conducted in Shebedino district of Sidama Zone, Southern Nations, Nationalities and People's Regional State (SNNPRS) of Ethiopia. Leku the capital city of the district is situated 290 km south of Addis Ababa and 27 km south of Hawassa. The study area shares common border with Hawassa town, Dale, Gueriche and Borecha Districts (SWAO, 2013). Shebedino district was selected on the basis of availability of sweet potato and suitability of agro-ecology for experimental plants and accessibility to the Hawassa University working facilities.

The District is 197.1 km^2 and 1800 to 2950 m. a. s. l. It receives 900 and 1500 mm rainfall annually between June-September and February-April. Mean temperature of the District ranges between 16 to 25°C. It has two agro-ecological zones: Dega in the range of 2500 to 2812 m. a. s. l (9.4% and has 4 Kebeles) and Weinadega in the range of 2000 to 2499 m.a.s.l. (90.4% and has 31 Kebeles), of these 3 Kebeles are in town and 32 in rural.

Intercropping plantation

Coffee plants which have similar age, number of coffee plants per 1200 m^2 on each farmer's land, relatively similar landscape, soil type and management for intercropping types (sole sweet potato, coffee and maize + haricot bean) were selected for intercropping. Four farmers were selected and an agreement was made with them for land preparation, plantation and management. A plot size of 1200 m^2 per farmer was sub divided in to three equal parts for successive plantation. The land was ploughed two times and vine seed of Hawassi-83 and Tula varieties were purchased for sequential plantation. Each variety under each farmer was planted on 200 m^2 of land first in 18 July, 2013. The second and third rounds were carried out on the same size plots at 15 days interval. Total planted area was 4800 m^2 (200 m^2 × 2 varieties × 3 planting interval × 4 farmers) was ploughed 3 times during growth period.

Farmers were selected to evaluate the biomass productivity and early maturity, viability and land cover based on their experiences of sweet potato production and using it as source of feed, food and income. The evaluators examined the fields three times (2nd, 4th and 6th month of growth). Fresh samples from each variety were collected from 1 m^2 at 4, 5 and 6 months of age (early, medium and maturity stages) for nutrient and biomass determination and partial budget analysis.

Chemical analysis

Dry matter and ash were determined according to AOAC (1990), nitrogen (N) content was determined by the Kjeldhal procedure and crude protein (CP) was calculated as N*6.25. Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) were analyzed according to Van Soest et al. *(*1991).

In vitro dry matter digestibility (IVDMD)

In vitro studies were conducted to estimate the potential digestibility of the pooled sub-samples of each of the 4 round forage cuts. The IVDMD was determined using the two stages *in vitro* Tilley and Terry procedure (1963) as modified by Van Soest and Robertson (1985).

Partial budget analysis

The three land types were assessed for comparative advantages based on samples taken on 100 m^2. All produces on the plots were recorded across the trial period. Income from coffee (non-processed beans), maize and haricot bean grains and byproducts and all costs (labor, seed, vine, tuber, etc) were estimated based on seasonal prices on local market. The price of vine and tuber were taken as constant values.

Partial budget analysis was conducted for measuring profit margin of intercropping (Upton, 1979). Net income (*NI*) was calculated as total return (TR) minus total variable cost *(TVC)*:

$$NI = TR - TVC$$

Change in net income *(ΔNI)* was calculated as change in total return *(ΔTR*) minus change in total variable cost *(ΔTVR):*

$$\Delta NI = \Delta TR - \Delta TVR$$

Marginal rate of return *(MRR)* as a measure of increase in net income *(ΔNI)* associated with each additional unit of expenditure *(ΔTVC)* was calculated as

$$MRR = (\Delta NI / \Delta TVC) \times 100$$

Data management and statistical analysis

Nutrient contents, biomass yield (DM and OM) and IVDMD of the morphological fractions and qualitative evaluation of the morphological fractions of the sweet potato varieties harvested from three intercropping at three stages of maturity were subjected to ANOVA using General Linear Model (GLM) procedure of SPSS Version 22 (SPSS, 2014). Productivity of the land used for sweet potato intercropping was subjected to descriptive statistics. Means were separated using Duncan's Multiple range test at $p<0.05$.

The model for analysis of variance was $Yijk = \mu + \alpha i + \theta j + \beta k + \alpha i\theta j + \alpha i\beta k + \theta j\beta k + \alpha i\theta j\beta k + eijk$ where: *Yijk* = DM and OM content/yield IVDMD (i=vine, leaf and stem with petiole); at three stages of maturity (j = 5th, 6th and 7th months of age of sweet potato) and four

Table 1. Dry matter and organic matter contents of morphological fractions (vine, leaf and stem-petiole) of the two varieties (Hawassi-83 and Tula) cut at three stage of growth.

Parameter	Stage of growth (months)	Morphological fractions						Overall mean	p- value
		FHV	FHL	FHSP	FTV	FTL	FTSP		
DM (g/kg)	4	139^{aC}	158^{aD}	142^{C}	124^{aB}	188^{E}	76^{A}	138±7	0.0000
	5	142^{abC}	162^{aD}	143^{C}	133^{bB}	190^{E}	79^{A}	142±6	0.0000
	6	149^{bC}	168^{bD}	143^{C}	133^{bB}	191^{E}	76^{A}	143±7	0.0000
	Mean	143^{C}	163^{D}	143^{C}	130^{B}	190^{E}	77^{A}	141±7	0.0000
	SEM	6	2	0	2	1	1		
	p-value	0.051	0.0080	0.422	0.0173	0.8464	0.1781		
OM (g/kg DM)	4	863^{aA}	860^{A}	862^{A}	853^{A}	915^{B}	863^{aA}	869±4	0.0000
	5	872^{bB}	855^{B}	863^{B}	835^{A}	913^{C}	853^{aB}	865±5	0.0000
	6	869^{bB}	858^{B}	863^{B}	833^{A}	913^{C}	848^{bB}	863±5	0.0000
	Mean	868^{B}	858^{B}	863^{B}	840^{B}	914^{A}	855^{B}	866±5	0.0000
	SEM	5	6	2	7	2	9		
	p-value	0.0096	0.2205	0.9063	0.0051	0.1250	0.0063		

Column means bearing different lowercase superscript letters are significantly different and row means bearing different uppercase superscript letters are significantly different ($p<0.05$). FHV, fresh Hawassi-83 vine; FHL, fresh Hawassi-83 leaf; FHSP, fresh Hawass-83i stem with petiole; FTV, fresh Tula vine; FTL, fresh Tula leaf; FTSP, fresh Tula stem with petiole; DM, dry matter; OM, organic matter, SEM, standard error of mean.

intercropping types (k= sole sweet potato, sweet potato intercropped with coffee and sweet potato intercropped with maize-haricot bean); μ = over all mean; αi = effect of morphological fractions on DM and OM content/yield and IVDMD; θj = effect of stage of maturity on DM and OM content/yield and IVDMD; βk = effect of intercropping on DM and OM content/yield and IVDMD; $\alpha i\theta j$ = interaction effect among the morphological fractions and three stages of maturity on DM and OM content/yield and IVDMD; $\alpha i\beta k$ = interaction effect among the morphological fractions and three intercropping types on DM and OM content/yield and IVDMD; $\alpha i\theta j\beta k$ = interaction effect among the morphological fractions, three stages of maturity and three intercropping types on DM and OM content/yield and IVDMD; *eijk* = random error

RESULTS

Nutrient content, biomass yield and *in vitro* dry mater digestibility of the sweet potato varieties

Dry matter and OM yield of morphological fractions cut at three stages of maturity are depicted in Table 1. The DM content of FHL and FTV at 3rd stage of growth was greater ($p<0.05$) than at 1st and 2nd; FTV having the least ($P<0.05$) at 1st stage of growth. The rest of the morphological fractions were similar in DM content at all stages of maturity. The OM content of all the morphological fractions of the two varieties cut at three stages of growth were similar ($p>0.05$).

The overall mean dry matter (DM) and organic matter (OM) yields of vines of the two sweet potato varieties intercropped with coffee and maize-haricot bean and cut at three stages of maturity is depicted in Table 2. Both DM and OM yields of vines of sweet potato intercropped with maize-haricot bean were highest of the other two intercropping ($p<0.05$).

The overall nutrient content of mixtures of the respective morphological fractions from the two sweet potato varieties under three intercropping and the three stage of maturity is depicted in Table 3. The DM content of FHL was greater than that of FHV and FHSP; and DM differences of morphological fractions of Tula were wide; FTSP had the lowest but FTL the highest. Leaf parts of the two sweet potato varieties had greater DM than vine and stem with petiole. The lowest OM content was in FTV but FTL had highest OM content of FTV and FTSP and all fractions of Hawassi-83.

The CP contents of FHSP were the least but the highest in FTL. EE content in both varieties of FHL, FTL and FTSP were the least but highest in FTV. The least NDF was in FTL but the highest was in FTSP. The ADF content of FTL was the least of all but that of FTSP was the highest. The least ADL was in FTL but the highest was in FHSP. The IVDMD of FHL and FTSP were similar and much lower than that of FHSP whereas FTL the FHV come in between.

Qualitative evaluation and partial budget analysis

As shown in Table 4 majority of evaluators/respondents ($p<0.05$) preferred Hawassi-83 to Tula in biomass productivity and yield, adaptability to intercropping, early maturity and marketability of the tuber. Farmers can reasonably evaluate crops according to the current local situation and benefits.

Table 2. Dry matter and organic matter yields of vines of the two sweet potato varieties produced under three intercropping types.

Yields	Intercropping types			SE	p
	Sole sweet potato	Sweet potato-coffee	Sweet potato-maize-haricot bean		
Dry matte (g/M²)	252[b]	174[a]	334[c]	22	0.0000
Organic matter (g/M^2)	217[b]	149[a]	286[c]	20	0.0000

Least square means within a row bearing different superscript letters are significantly different.

Table 3. The pooled nutrient content and *in vitro* DM digestibility (Mean ±SE) of the morphological fractions of Hawassi-83 and Tula harvested at three stages of maturity from three intercropping types.

Nutrient (g/kg DM)	Morphological fractions of sweet potato						Overall mean	p-value
	FHV	FHL	FHSP	FTV	FTL	FTSP		
DM (g/kg)	143±20[c]	163±7[d]	143± 1[c]	130±7[b]	190±6[e]	77±5[a]	141±2	0.0000
Ash	133±14[b]	141±17[c]	138±3[c]	160±23[d]	85±2[a]	144±28[c]	134±3	0.0000
CP	176±1[c]	243±0[de]	113±1[a]	222±1[d]	269±1[e]	134±0[b]	193±3	0.0000
EE	127±2[c]	83±0[a]	124±3[c]	152±0[d]	99±1[b]	85±0[a]	112±2	0.0000
NDF	345±32[c]	311±26[b]	349±0[c]	350±20[c]	276±47[a]	416±1[d]	341±2	0.0000
ADF	207±4[c]	169±1[b]	249±1[d]	215±2[c]	139±3[a]	290±3[e]	212±2	0.0000
ADL	170±0[c]	114±0[b]	193±2[d]	171±1[c]	99±3[a]	183±3[d]	155±2	0.0000
Digestibility (g/kg DM)								
IVDMD	809±1[b]	780±3[a]	843±6[c]	824±0[bc]	843±0[c]	770±1[a]	812±2	0.0000

DM, dry matter; CP, crude protein; EE, ether extract; NDF, neutral detergent factor; ADF, acid detergent factor; ADL, acid detergent lignin; IVDMD, *in vitro* dry matter digestibility; FHV, fresh Hawassi-83 vine; FHL, fresh Hawassi-83 leaf; FHSP, fresh Hawassi-83 stem with petiole; FTV, fresh Tula vine; FTL, fresh Tula leaf; FTSP, fresh Tula stem with petiole.

Table 4. Qualitative evaluation of Hawassi-83 and Tula of the respondents in the study area (N=10).

Parameter	Values			Test	
	Hawassi-83 N (%)	Tula N (%)	Total N (%)	χ2	*p-value*
Biomass (vine and tuber) yield	9 (90)[b]	1(10)[a]	10(100)	10	0.002
Better yielder in inter-cropping	7(70)	3(30)	10(100)	10	0.002
Tuber palatability	9(90)[b]	1(10)[a]	10(100)	10	0.002
Short maturation time	8(80)	2(20)	10(100)	10	0.002
Marketability	7(70)	3(100)	10(100)	10	0.002
Overall	40(78)[b]	10(22)[a]	50(100)	50	0.000

Row values with different superscript letters are significantly different. N= number of respondents.

The total revenue obtained after six months of the trial from different intercropping is depicted in Table 5. The products from sole sweet potato cultivation are vine and tuber but from the intercropping coffee, maize grain and haricot bean are also included. Sole plantation or intercropping of Hawassi-83 was more profitable than those of Tula. Sweet potato intercropping with maize-haricot bean was more profitable than with coffee.

DISCUSSION

The studies of Tesfaye et al. (2011) have shown that the DM content of unspecified sweet potato variety was 92% but neither of the DM contents of vines from the varieties in this study agreed with this value. Netsanet (2006) reported DM of unspecified variety of sweet potato vine, leaf and stem were 21.05, 21.7 and 18.67%, respectively but only the leaf part was close to the DM content of FTL. The study of Tesfaye and Amenti (2008) on Hawassi-83 vine was lower in DM content than result obtained in this study, which may be attributed to differences in stage of maturity, types of treatment and soil type.

The DM of FTSP was the least but that of FTL the highest. Leaves of the two sweet potato varieties harvested at third stages of growth varied in DM content

Table 5. Productivity of the two sweet potato varieties intercropped on farmers' land (800 M^2) during June- March, 2013

Parameters	Intercropping type																	
	Sole sweet potato				Sweet potato-coffee						Sweet potato-maize-haricot bean							
Variety	Hawassi-83		Tula		Hawassi-83			Tula			Hawassi-83				Tula			
Product type	Vine	Tuber	Vine	Tuber	Vine	Tuber	Coffee	Vine	Tuber	Coffee	Vine	Tuber	Maize	HB	Vine	Tuber	Maize	HB
Fresh (kg)	445	960	286	182	288	360	45	218	69	50	653	216	224	80	308	142	224	132
Cost (Birr/kg)	667	3840	429	728	432	1440	227	327	277	252	980	864	1343	480	462	568	1344	795
Sums of revenue (Birr)	4508		1157		2099			856			3667				3169			
Return due to intercropping (Birr/M^2)	5.63		1.44		2.62			1.07			4.58				3.96			
Return due to intercropping (Birr/M^2)	3.53				1.84						4.27							

with no visible trend in the rest of the fractions; while similar trend was observed in DM yield between FTV and FHV at last stage of growth; the rest of the fractions showed no differences. The highest DM and OM yields were obtained from sweet potato maize-haricot bean intercropping.

The OM of FTV and FTSP was the least but that of FTL the highest. All morphological fractions within a species showed no difference in OM in the three stage of growth. The OM content of vine and leaf earlier reported (Netsanet, 2006) nearly agreed with the results of this study but not of the stem. The study of Tesfaye and Amenti (2008) on Hawassi-83 vine was lower in OM content than this study but nearly agreed with OM content of FTV and Hawassi-83 with Kudade in their study.

The CP was the highest but EE the lowest in FHL and FTL. The CP and EE contents were inversely related in the morphological fractions. The CP content of FHSP and FTSP were the least compared to vines and leaves which agrees with earlier results reported (Etela and Oji, 2009). The CP content of FHL and FTL were the highest compared to vine and stem with petiole in both varieties and agrees with earlier results (Etela and Oji, 2009; Netsanet, 2006; Orodho et al., 1993) but disagrees with that of Tesfaye et al. (2011). The CP contents of FHL and FTL were greater than 20% which agrees with the study of Orodho et al. (1993) it was higher than the finding (20.9% CP on DM basis) of Woolfe (1992) stated for sweet potato green. The CP content of the vine was greater than CP content of all other parts (FAO, 1994). Tesfaye and Amenti (2008) reported higher CP content of Hawassi-83 vine than that of FHL but nearly agreed with the CP content of Falaba Damota which could be attributed to differences in parts examined, soil type, stage of maturity and sweet potato varieties.

The NDF value of FTSP agreed with the NDF value reported of sweet potato stem (Netsanet 2006) but disagreed with that of Etela and Oji (2009) in which leaf blade had the highest NDF. The study of Tesfaye and Amenti (2008) on Hawassi-83 vine was lower than that of FHV and FTV of this study. The NDF content of FTL nearly agrees with that of Hawassi-83; FHL with that of Koka-12 and FTV with that of Belela.

The ADF content of FTL was the least of all but that of FTSP was the highest which generally disagrees with earlier report (Netsanet, 2006). The study of Tesfaye and Amenti (2008) on Hawassi-83 vine was higher in ADF content than FHV in this study but it nearly agreed with Halaba and Damote; and that of FHSP in this study agrees with Koka-12 and FTSP with Bereda and Belela. The ADL content of sweet potato stem earlier reported (Netsanet, 2006) agrees with the results of this study. None of the ADL contents in this study agree with the findings of Tesfaye et al. (2011).

The least CP and higher ADL contents in FHL are possible reasons for reduced digestibly but in the FTL the higher CP and least ADL contents must have been in favor of the highest digestibility rate. As the ADL content increased in Hawassi-83, digestibility increased but the opposite happened in Tula. In Hawassi-83, as the CP content increased, the digestibility decreased but the opposite happened in Tula parts. The FHSP was more digestible than FHV but FTL was more digestible than FTV. Vines in both varieties had lower IVDMD which agrees with the results of an earlier study (Netsanet, 2006).

The highest DM, CP and IVDMD and lowest NDF, ADF and ADL were from FTL but medium DM, highest CP and lowest ADL and IVDMD were from FHL. The low ADL in FHL suppressed

IVDMD but in FTL it favored IVDMD however in FHSP, the ADL and IVDMD were linearly related.

Hawassi-83-maize-haricot bean intercropping gave better land productivity, vine biomass and total revenue; possibly due to improved soil fertility by nitrogen fixation of haricot bean and thus to overcome the critical land and feed shortage and reduced land productivity of the district and region, intercropping sweet potato with maize-haricot bean should be promoted.

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Conflict of Interests

The authors have not declared any conflict of interests.

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Improving livestock productivity: Assessment of feed resources and livestock management practices in Sudan-Savanna zones of West Africa

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Evaluation of existing and potential feed resources was conducted in Orodora district in the Southern region of Burkina Faso using Feed Assessment Tool (FEAST). The assessment was carried out through focus group discussions and individual interviews in Mahon and Sayaga communities in Orodara district. The study sites were characterized by mixed crop-livestock production systems. Seventy-percent of cropping activities were focused on fruit tree cultivation while food crop production accounted for the rest. Livestock species (predominantly local breeds) in the area included cattle sheep, goat, pig, poultry and donkey which are kept for different purposes. The main source of household income is crop production while livestock production contributed 35 and 45% to the household income in Mahon and Sayaga, respectively. In both study sites, natural grazing contributes highest (49 and 64% respectively) to the dry matter (DM) content of the total diet. Cultivated fodder contributed 1% of dry matter (DM), metabolizable energy and crude protein to the total diet of the existing feed resource in Mahon, while farmers in Sayaga depended more on purchased feed than in Mahon. Constraints to livestock production in the study sites included shortage of water in the dry season, insufficient quantity and quality of feed in the late dry season of the year and high cost of veterinary drugs and services. To mitigate these constraints farmers suggested an integrated approach to improve livestock production through: construction of small reservoir to provide water for human and animal consumption; training on the integration of forage legume into both tree and arable cropping, and efficient utilization of available feed resources; establishment of a veterinary service and drug centre in the village; better management of the existing water resources.

Key words: Animal nutrition, livestock productivity, feed resources, feed scarcity.

INTRODUCTION

Livestock sector is the second most important source of income in Burkina Faso, and its contribution to the country's gross domestic product (GDP) was estimated at 15% (Nianogo and Thomas, 2004) and varies between 18.3 and 19.5% over the period 2001 to 2008 (MRA, 2011). Nineteen percent of Burkina Faso's export

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products are from livestock production (MRA, 2004). The population of ruminant livestock species of Burkina Faso is estimated to be 8.7, 11.4 and 7.3 million heads of cattle, goats and sheep, respectively (FAO, 2005) and the annual growth rate was reported to be 4.7, 3.3 and 2.3% for goats, sheep and cattle, respectively (MRA, 2004). Most of these animals are found in the arid and semi-arid part of the country. However, the livestock numbers are growing faster in the sub-humid zone (LEAD, 2005), as a result of the recent pattern of transhumance in West Africa in which pastoralists and their livestock transited from the more arid Sahelian region in the north to the more humid Sudano-Guinean regions in the south (Ayantunde et al., 2014). The extensive production system using low inputs is common in the country (MRA, 2004), while the "mixed system" in which crop and livestock production are jointly practiced is the most dominant in the sub-humid zone (Seré and Steinfeld, 1996).

Nutrition of cattle, sheep and goats in the agro- pastoral systems of the Sahelian and Sudanian-Savanna zones of West Africa is essentially based on the exploitation of naturally occurring herbaceous and ligneous plant species, and crop residues. Qualitative and quantitative forage shortage, particularly in the dry season, is the major constraint in these farming systems (Breman and de Ridder, 1991). During periods of feed shortage (the dry season), livestock owners often use agricultural by-products such as cereal (maize, millet, sorghum, wheat) bran, cotton seed cake in the cotton producing zone, cowpea hay and groundnut haulm to supplement animal feed. Herders and others pastoral groups sometimes cut down branches from various tree species such as Acacia spp., *Balanites aegyptiaca, Pterocarpus lucens* and *Boscia angustifolia* to feed their animals during the dry season (Gautier et al., 2005).

Pastures in Burkina Faso are subject to seasonal variability. Generally, there is good availability of forages in terms of quantity as well as quality in the rainy season which lasts for about 6 months (May to October) in the sub-humid zone. This is followed by another 6 months of dry season (November to April) when there is a rapid decline in forage resources in both quantity and quantity. This has been considered as the major limitation to meeting the nutrient requirements of grazing livestock for most of the year (Kavana et al., 2007). With this constraint, animals that rely on natural pastures as basal diet cannot perform at high level without feed supplementation which may be expensive for low income farmers (Mohammed-Saleem, 1990).

Overcoming the constraint of feed scarcity requires a well-targeted assessment of available feed resources as to inform and guide interventions to address the problem. In this regard, a systematic and rapid methodology for assessing feed resources at site level with a view to developing a site-specific strategy for improving feed supply and utilization through technical and organizational interventions has been developed by International Livestock Research Institute (Duncan et al., 2012). The objective of this study was to evaluate existing and potential feed resources in the study sites to guide feed-related interventions to improve livestock productivity in the dryland areas of Burkina Faso.

MATERIALS AND METHODS

Description of study site

This study was conducted in Mahon and Sayaga communities in Orodara district in the southern part of Burkina Faso within latitude 11.3167°N and longitude 4.5667°W (Plate 1). Orodara district is in the South Sudan agro ecological zone of Burkina Faso with one rainy season per year and annual total rainfall ranging from about 900 to 1200 mm. The main textural classes are loamy sand, sandy loam, loam, sandy clay loam, clay and silty clay loam. These characteristics impose some limitations on crop production including restriction of plant rooting systems, high runoff, and low retention of water and nutrients (FAO, 2001). Mixed crop-livestock system is the dominant agricultural production system in the study site. The dominant crops grown in the study site include cotton, yams and cereals (sorghum, millet and maize). In addition, fruit trees such as mango, citrus and cashew are widely cultivated in the study area. Livestock are owned and managed under traditional production systems which are characterized by low productivity in terms of milk and meat (Figure 1).

Methodologies and implementation of the survey

Evaluation of existing and potential feed resources was conducted using Feed Assessment Tool (FEAST) developed by International Livestock Research Institute (ILRI) (Duncan et al., 2012). FEAST is a systematic method used to assess local feed resource availability and use. It helps in the design of intervention strategies aiming to optimize feed utilization and animal production. It comprises Participatory Rural Appraisal (PRA) using semi-structured questionnaire in a focus group discussion and individual interviews to collect both qualitative and quantitative data. The study was conducted from April to May 2014.

Twenty-six farmers which comprised 18 men and 8 women in Mahon and 20 farmers (15 men and 5 women) in Sayaga were selected as representatives of the community to participate in group discussions using the participatory rural appraisal (PRA) approach to provide an overview of the farming systems and to identify constraints and opportunities for improving livestock production in the 2 study communities. For the individual interview, twelve farmers were then selected representing 3 wealth categories within the community namely average, above average and below average in terms of land area owned and number of livestock possessed by the household. Four farmers from each wealth category were individually interviewed to collect quantitative and qualitative data on feed resources in the community. Through the use of a structured questionnaire, farmers were asked to identify and describe their perception of quality of the available feed resources and then rank them according to their indicators for feed quality. Samples of available feed resources offered to the animals were collected and analyzed for nitrogen, ash content, fibre components (NDF, ADF and ADL) and *in vitro* organic matter digestibility.

Data analysis

The quantitative data collected from individual key informant

Figure 1. Map of Burkina Faso showing the study site.

farmers were entered into the FEAST Excel template (www.ilri.org/feast) and analyzed. FEAST qualitative data collected were examined and summarized for each major topic and linked with FEAST individual interview output. Results are presented in tables and figures in the results and discussion of this work.

RESULTS AND DISCUSSION

Overview of the farming system

From the results of the survey, farming systems in both Mahon and Sayaga can be described as mixed crop-livestock production with fruit trees dominating the cropping systems. The major crops grown in both areas were maize, millet, sorghum, groundnut and cowpea which were mainly grown during the raining season. Other crops such as cabbage, carrots and other vegetables are planted in the dry season both for consumption and income. Farmers in both Mahon and Sayaga described two distinct cropping seasons based on the rainfall patterns and the time of crop harvest. The long rainfall season locally called 'Gnigboke' extends from May to October, while the dry season 'Gnigbeke' is from November to April.

Crop production in the study sites was dominated by fruit tree crop. Farmers confirmed that 70% of their cropping activities were focused on fruit tree cultivation (arboriculture) while food crop production accounted for the rest. The fruit tree crops were mango (which is 60% of the production), orange and cashew. Both fruit trees and arable crop production were rain-fed. According to Badini et al. (1997), 75% of the crop production in Burkina Faso which is the main source of food and income for the majority of people is rainfed. Maize was the dominant arable crop in both Mahon and Sayaga followed by sorghum (Figures 2 and 3). However, rice was considered as one of the arable crops in Mahon due to the presence of lowland areas which could support rice cultivation.

Availability of irrigation facility is a great constraint to crop production in the study sites due to the absence of perennial rivers. The only dam available is about 4 km to Mahon making it difficult to practice irrigation and this limits the productive ability of farmers in the dry season

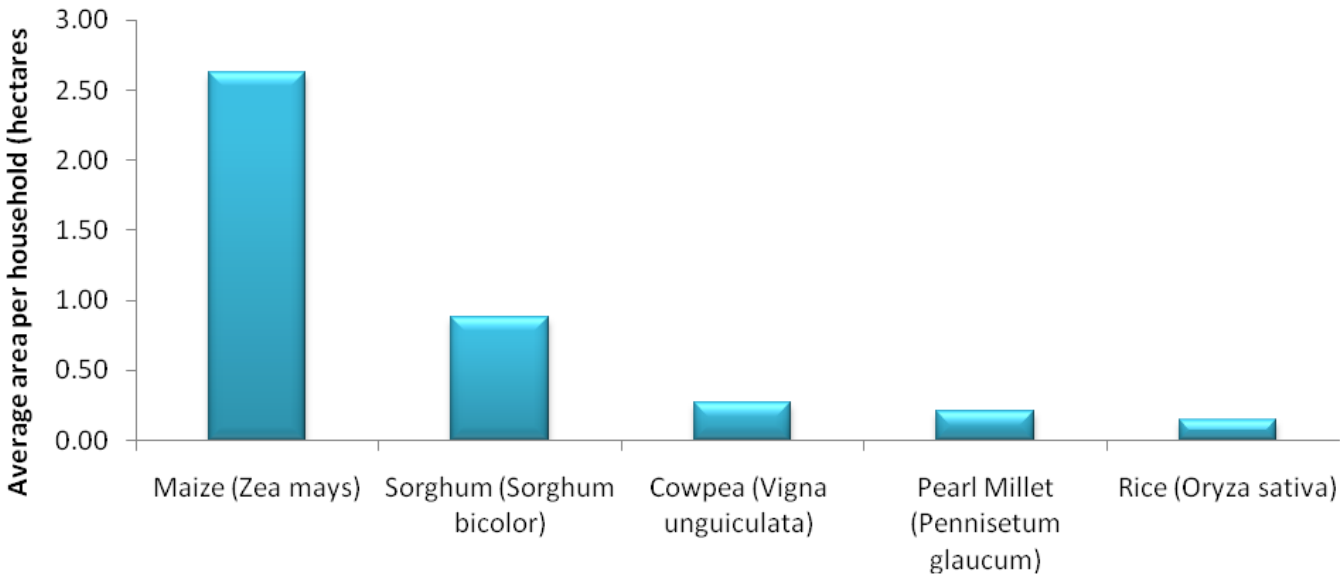

Figure 2. Average area (ha) cultivated per household of dominant arable crops in Mahon.

and animals often trekked long distance in search of water. Farming at this time of the year (dry season) in Mahon is also limited to vegetables, carrots, cabbage and tomatoes which are planted on small areas in lowland where the soil water is still available in sufficient amount to grow such crops. However, farmers reported that due to erratic rainfall pattern, there is reduction in the level of the water tables in the lowlands in the last 2 years as a result of low rainfall. There are no low land areas in Sayaga, therefore cropping activities were limited to the raining season.

Both household and hired labours for agricultural activities are available all year round in Mahon and Sayaga. Household members serve as labour force for certain farm activity such as fertilizer application. However, since most household members have their own farms to cultivate and may not always be available and adequate to undertake farm activities such as weeding and harvest, hired labours are therefore required at such times. In Mahon, there is an organized labour group where farmers can go and hire for the cropping activities. The cost of hiring group labour ranged from 10,000 to 25,000 CFA per day (21 to 53 USD as at the time of the study) which sometimes consisted of 15 to 20 men per group. A male farm labourer is hired for an average of 1500 CFA per day (3 USD) while female labourer could be less than 1000 CFA per day (2 USD) depending on the nature of work. Women are mostly needed during harvest and post-harvest operations. However in Sayaga, labour cost does not depend on the nature of farm activities.

Access to credit was described by the farmers as another constraint to agricultural production in the study areas. Although there is a local credit bank for farmers owned by the government (Caisse populaire), access to credit is quite difficult. Kuwornu et al. (2012) reported similar situation from a study conducted in Ghana. The main bottleneck as noted by the farmers in the study areas was the long process of applying for and receiving the credit. Farmers reported that it could take about 8 months before it is released. Sometimes, the loan is released after farming activities had ended for the cropping season. Apart from the problem of timing, farmers do not have the required collateral needed as guarantee to access credit. Another issue with access to credit according to the farmers interviewed was that available credit is mostly given for crop production for the purchase of farm inputs like fertilizer and improved seeds, and to pay hired labour which implies that it is difficult to access credit for livestock production. According to the respondents, there is no credit facility available for the farmers in Sayaga for both crop and animal production, although famers in the area were willing to organize their own cooperative if they could be trained on how to establish and run it.

Inputs such as fertilizers, improved seeds and tractor services are not readily available according to the respondents. Farmers also reported that some input such as fertilizer and seed are sometimes of low quality. At present, farmers in Mahon set up a community seed store where each farmer contributes his own grain seed which they later share during planting. Farmers in Sayaga reported that most agricultural inputs are not available in local markets, fertilizer prices were reported to be too high while other inputs are in short supply compared to Mahon. This could be attributed to the far distance of Sayaga to the major town in the region.

According to the farmers interviewed, an average of 8%

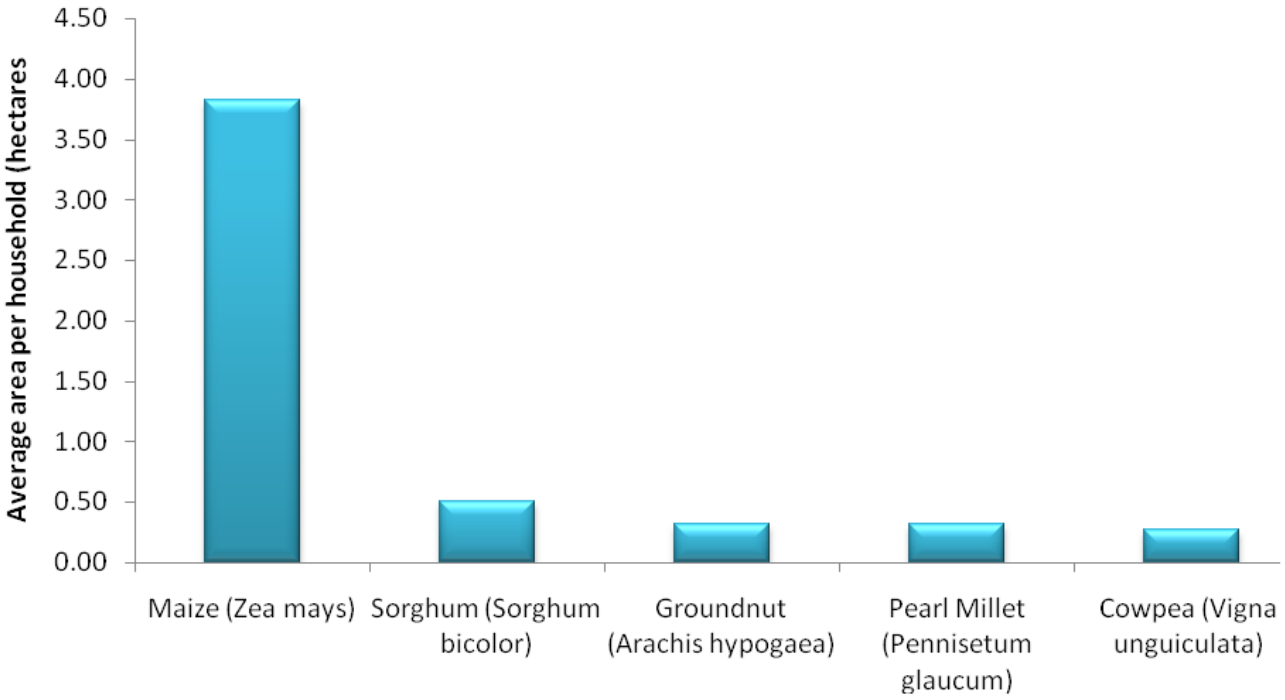

Figure 3. Average area (ha) cultivated per household of dominant arable crops in Sayaga.

of the people in every household migrated from Mahon to the cities and towns in search of better paid jobs. These groups of people are mostly youth and middle aged population. Few of them return to the village during the festive periods or at the end of each year. Migration out of Sayaga is quite high as about 25% of people from each household had moved out to cities and towns in search of better livelihood opportunities. Higher migration in Sayaga could be attributed to its remoteness. Deshingka (2010) reported higher incidence of migration among chronically poor people living in remote villages.

Household characteristics, land holding and land use pattern

Maholn is made up of approximately 180 households with an average of 20 to 25 people in each household. The average land holding in the study sites was between 5 to 10 ha for different wealth categories (Figure 3). The results indicated that the majority of the farming households are smallholders with land sizes less than 5 ha. Few households classified as medium and large farmers had few household size.

The farmers remarked that in the past, farm lands were jointly owned and cultivated by every member of the family; some 20 years ago, the lands were divided within the family to every member. As a result of land fragmentation, arable land is in short supply as the family size continues to increase and there is no more fallow practice in the communities. This fragmentation of limited farmland leads to shorter fallow periods and in some cases continuous cultivation (Bezuayehu et al., 2002). Continuous cropping significantly of the same land affects soil physical and chemical properties by increasing soil bulk density and reducing both macroporosity and macro-aggregates, resulting in less water and nutrient availability (Qin et al., 2007). Consequently, crop yields become unstable and tend to decline, especially in low rainfall years (Sainju et al., 2009). The 2% landless farmers in Mahon were the Fulani (pastoralist) migrants whose major livelihood strategy is animal production and do not have right to own land in the community.

Sayaga consist of about 110 household with the average family size of 16 people per household. The farm land varied among the households but average farm size was about 5 ha. Majority of the households fell into the category of smallholder farmers with 1 to 5 ha of land (Figure 4), followed by the medium land holding and large land holding farmers. Namubiru-Mwaura and Place (2013) reported average size of 3 ha and above as land owned by smallholder farmers in West Africa countries namely Burkina Faso, Mali and Niger. There was no landless farmer in Sayaga (Figure 5).

Major sources of household income

As presented in Figure 6, the main sources of household income were crop farming, livestock production and other

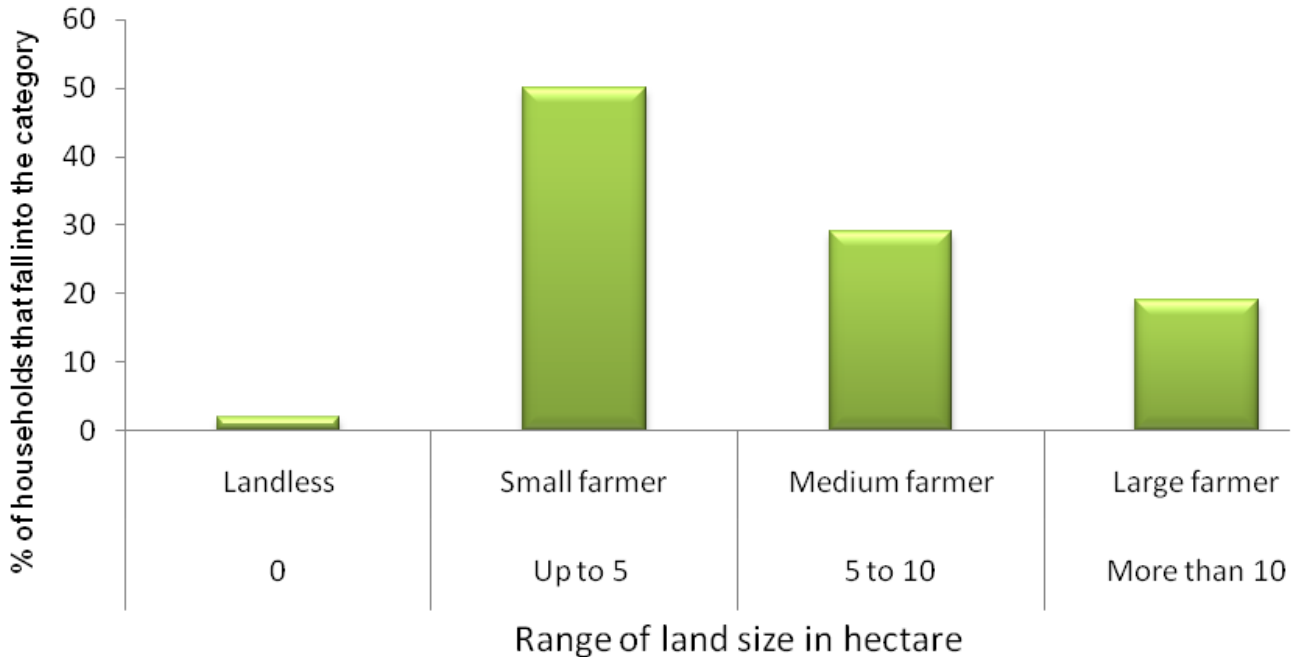

Figure 4. Distribution of land area cultivated for households in different wealth categories in Mahon.

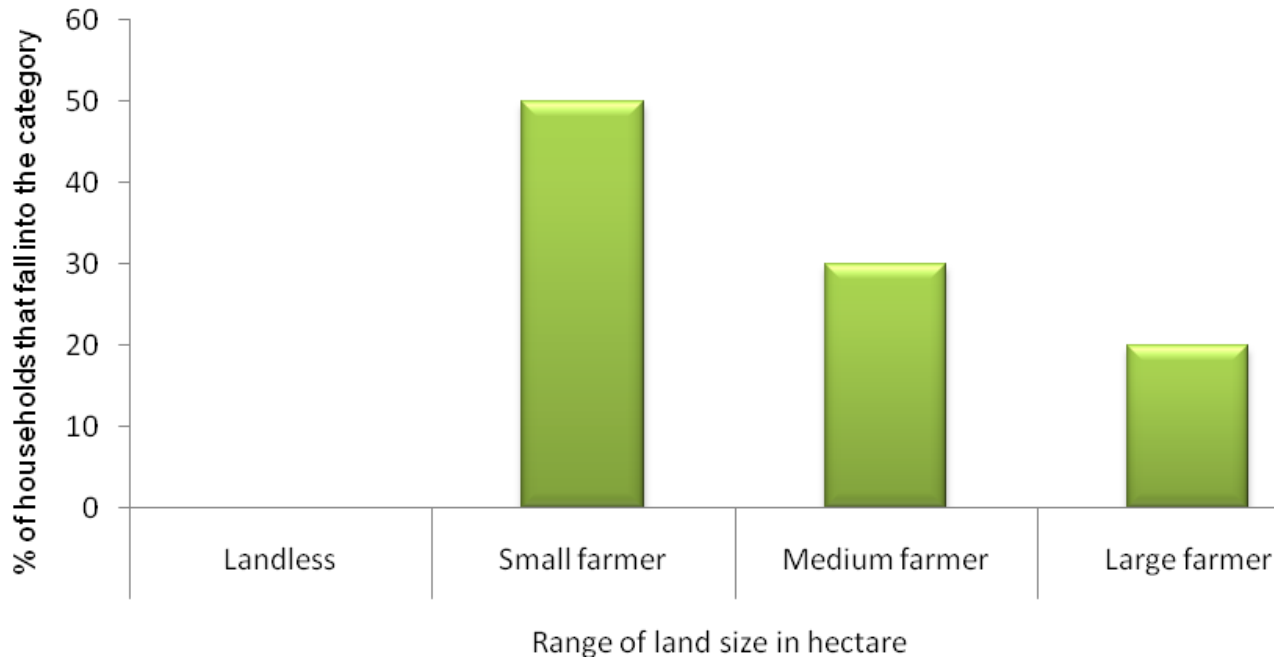

Figure 5. Distribution of land area cultivated for households in different wealth categories in Sayaga.

occasional businesses. Basically, income generated from crop production comes mainly from fruit trees and arable crops. Livestock production also contributed 35 and 45% to the household income in Mahon and Sayaga, respectively. This comes from sales of milk and live animals in general. This confirmed the report of Mabel et al. (2010) that livestock contributes a large proportion of the income of smallholder farmers in sub-Saharan Africa. The farmers' responses showed that non-farm businesses were also important sources of income in

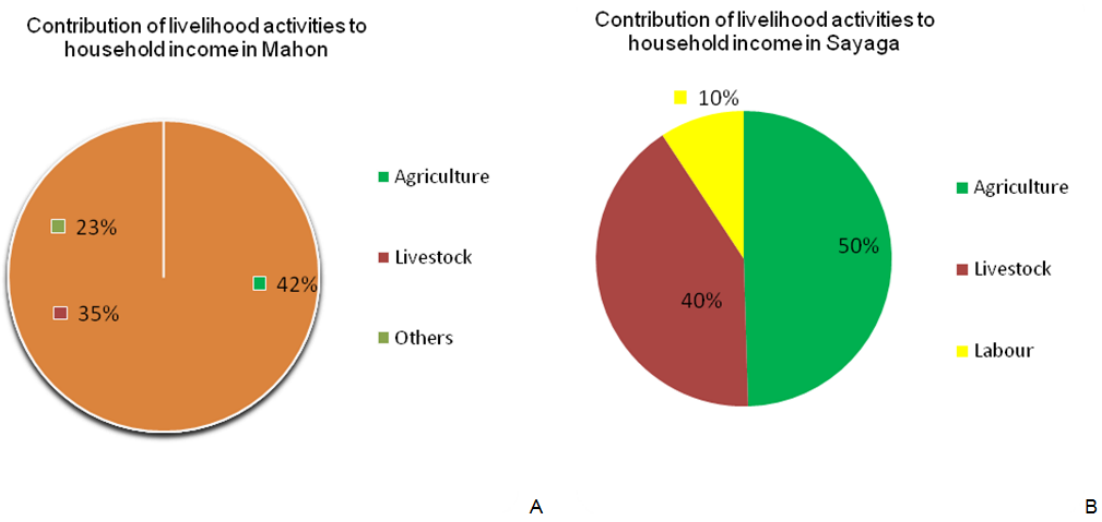

Figure 6. Contribution of livelihood activities to household income in Mahon and Sayaga.

Mahon and Sayaga. The result showed that crop and livestock production contribute more to household income in Sayaga than Mahon. On the other, household income derived from other sources such as off-farm businesses was higher in Mahon (23%) than (10%) in Sayaga. The distance and poor access road to nearby town may be responsible for this. Mahon is located 25 km from Orodara a semi-urban town with commercial activities which may influence the ease of engaging in off-farm businesses for farmers in Mahon.

In general, the direct agricultural income of the poor is not enough to sustain their livelihoods, either because of landlessness or because the land they own or lease is insufficient. Furthermore, wage employment in agriculture is highly seasonal, therefore the poor farmer value non-farm sources as means of earning additional income (IFAD, 2002).

Livestock assets and roles

Livestock form an integral part of agriculture and almost every farming household keep ruminants and/or indigenous chicken and different livestock species were kept for various purposes. Livestock species in the area included cattle, sheep, goat, pig, poultry and donkey. There were no improved breed of cattle or other animals in both Sayaga and Mahon. The results of this study indicated that cattle are the dominant livestock species in the areas (Figures 7 and 8). Farmers reported that approximately 70 and 65% of the households keep local cattle breeds and draught cattle with an average number of six and four per household in Mahon and Sayaga, respectively. Generally, cattle are kept for the purpose of draft power, manure, milk and cash income. According to farmers, draft cattle are the most important animals because of their use for cultivating cropland, crop threshing and straw transportation. Peeling and Holden (2004) had stated that cattle in the communal areas have multiple uses that include acting as a source of draught power, transport, milk, manure and savings among others. About 90% of the households owned sheep with the average number of eleven per household. They are mostly used as source of cash and meat. From the survey, every household in Mahon reared at least 10 local chickens. These chickens are sold as need arises, and are also slaughtered for consumption during festivals or ceremonies but occasionally for food. Although the number of donkey per household is very few, about 80% of the households in Mahon have at least one donkey. The primary purpose is for transportation of goods, crop harvests, people and fetching of water.

Livestock housing and management systems

In the study sites, different housing and feeding management practices were used for different livestock species and different classes of animals. Cattle and donkeys were housed in the shed made of wooden poles and thatched roof. Due to their vulnerability to diseases, sheep and goats are provided better housing near the homestead during the raining season to protect them from diseases. During the rainy season, lactating cows are also housed in the shed and they are often released for grazing. No housing system is practiced for pigs and poultry. The primary feeding practice in the study areas was open grazing. Herding was done when the number of animals is about ten and above. When the number of animal is few, in particular one or two, they are

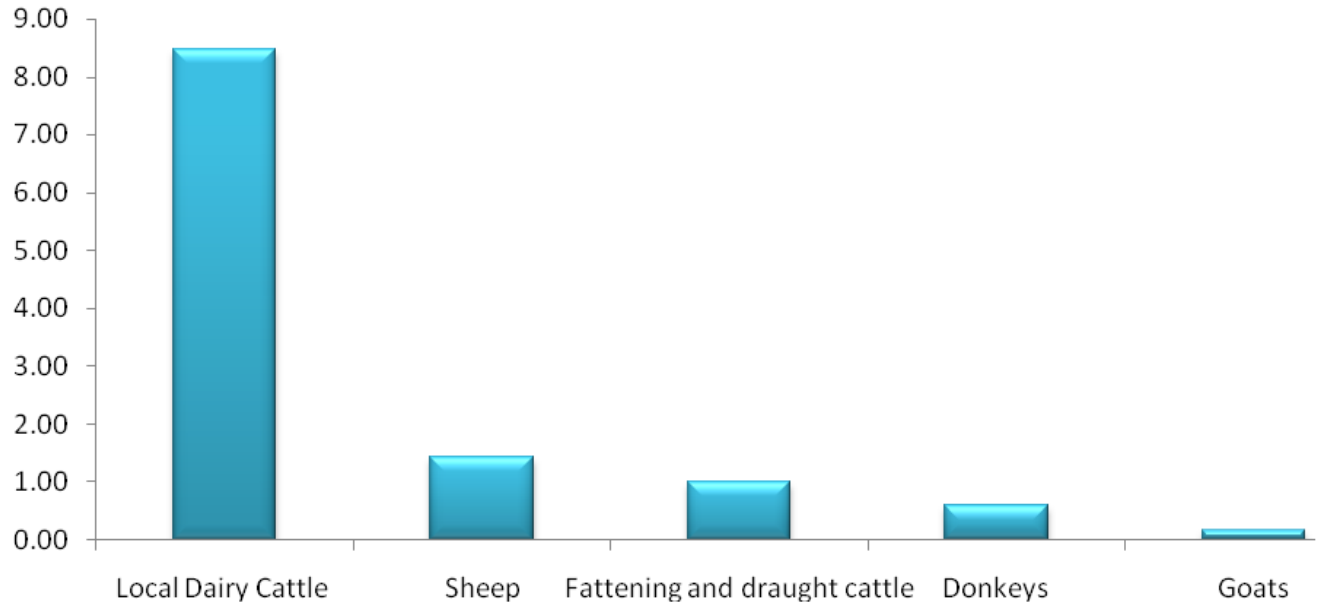

Figure 7. Average livestock holdings per household of dominant species (Tropical Livestock Unit) in Mahon.

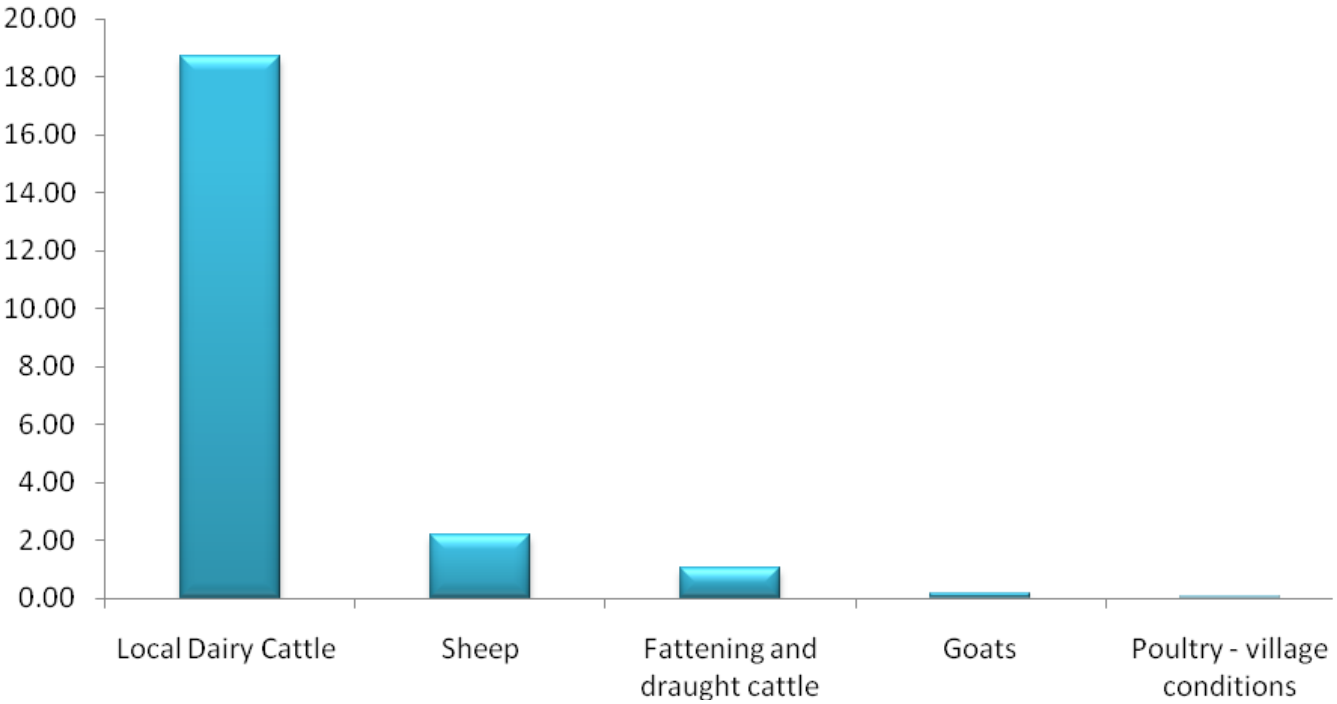

Figure 8. Average livestock holdings per household of dominant species (Tropical Livestock Unit) in Sayaga.

usually tethered.

Generally, farmers tethered their animals and feed them with crop residues. Occasionally, feed concentrate is offered during the dry season when there are limited feed resources. During the raining season, which is also the cropping time, open grazing is almost restricted to keep the animal from damaging crops. However, animals are allowed to graze near the homestead. According to the farmers, residues from cereals (maize, sorghum and millet) are chopped before being offered as feed, and

sometimes salt is sprinkled to add taste to some dry residues. One particular farmer in Mahon had practiced urea treatment and noticed a remarkable improvement in his animal performance over time. However, he discontinued for the fear of urea toxicity as a result of inappropriate dosage and financial constraint. Urea treatment of crop residues was not practiced in Sayaga due to cost. The farmers were aware of the potentials of such treatment to increase feed intake and animal performance, yet the costs of procuring it discouraged its use.

Results from another study in Zimbabwe revealed that labour availability, training, access to urea fertilizer and markets, and extension services are the main factors affecting adoption of urea treatment of maize stover technology (Mudzengi et al., 2014). Fodder production was not common in the study areas. Poor access to seeds, the problem of land rights and scarcity of land had been identified as major obstacles to adoption of fodder production in Burkina Faso (Kagone, 2006). However, one of the respondents in Mahon reported planting about 0.06 has of *Mucuna pruiens* over a period of 2 years and occasionally planted fodder maize but could no longer continue because of shortage of land and inability to harvest and conserve the seed of Mucuna.

Veterinary and artificial insemination (AI) services

Veterinary services were available to the farmers in both villages but not easily accessible. The veterinary clinic is located at about 8 km from Mahon and 15 km from Sayaga. The distance and the cost of transportation which is about 1500 FCFA (3 USD) make the service difficult to access. Farmers reported that about 2000 FCFA (4 USD) used to be spent to bring the veterinarian to Mahon for general treatments. In Sayaga, veterinary officers seldom visit the village but the farmers often invited them when it becomes inevitable which costs about 5000 FCFA (10 USD) due to long distance. Farmers described different symptoms of diseased condition in cattle such as fever, weakness and loss of appetite. Others are lameness and mange which cost 400 FCFA (0.8 USD) per cow to treat. The farmers also reported a disease condition in their cattle, whereby the animals are eating stones, sticks or plastic bags. Based on the symptoms described by farmer, this condition may probably be allotrophagia, although no clinical diagnosis was carried out. Allotrophagia had been reported as a diet-related disease of depraved or perverted appetite, whereby the animals start eating objects that they normally do not eat (Anderson, 1994; Akgul et al., 2000). This condition is probably due to mineral deficiency in the available feeds resources, natural pasture and crop residues, in the study sites. AI services were not available in the study sites. The practice is not common in the area according to the respondents. Uncontrolled mating with existing local bulls was the common practice in both study communities. In Sayaga, 20% of the farmers recalled being aware of AI but have never seen it practiced. However, they are willing to adopt AI if they can be trained and have access to it.

Major livestock feed resources and seasonal availability

The major feed resources were natural pasture and crop residues (Figures 9 and 10). Naturally occurring green fodder materials such as weeds from cropping areas, roadsides and grasses also served as sources of feeds particularly at the onset of rains. However, its availability was generally low in November to April to June which are the period of dry season. Crop residues become available in October after the crop harvest while availability of green forage declines. This may contribute to the increase in the quantity of commercially formulated ration purchased by the farmers as the dry season prolongs. Since the majority of the cultivated land area is allocated to cereal crops production, crop residues become the major feed resources during the dry season. In Sayaga, cereal crop residues (maize and sorghum stovers) and legumes (groundnut and cowpea haulms) formed parts of available feed resources (Figure 10). The collected residues are stacked in piles near homesteads and animals were given small quantities in the morning and evening.

The result showed that the quantity of collected crop residues used as livestock feed was higher in Sayaga than in Mahon. According to farmers, greater percentages of the crop residues were left on the field. These are also grazed along with naturally occurring forages which made grazing a major feeding method in both study sites. The result also showed that farmers in Mahon use commercially formulated ration as feed resource throughout the year while in Sayaga, farmers fed commercially formulated ration during the dry season till the onset of rains (Figures 9 and 10).

Purchased feed

The type, quantity and prices of purchased feed resources at the time of the study as described by the farmers were presented in Table 1. All the farmers interviewed in Mahon purchased supplementary feeds in the last 12 month to the time of the study. The available purchased feeds as reported by the farmers were maize bran, cotton seed cake, cowpea hay, *Parkia biglobosa* (pulp) leaves and commercially mixed ration (Table 1). *P. biglobosa* is one of the dominant tree species in study sites. The seed is used in making a local condiment. Ninety percent of the respondents purchased significant amounts of *P. biglobosa* pulp which was one of the

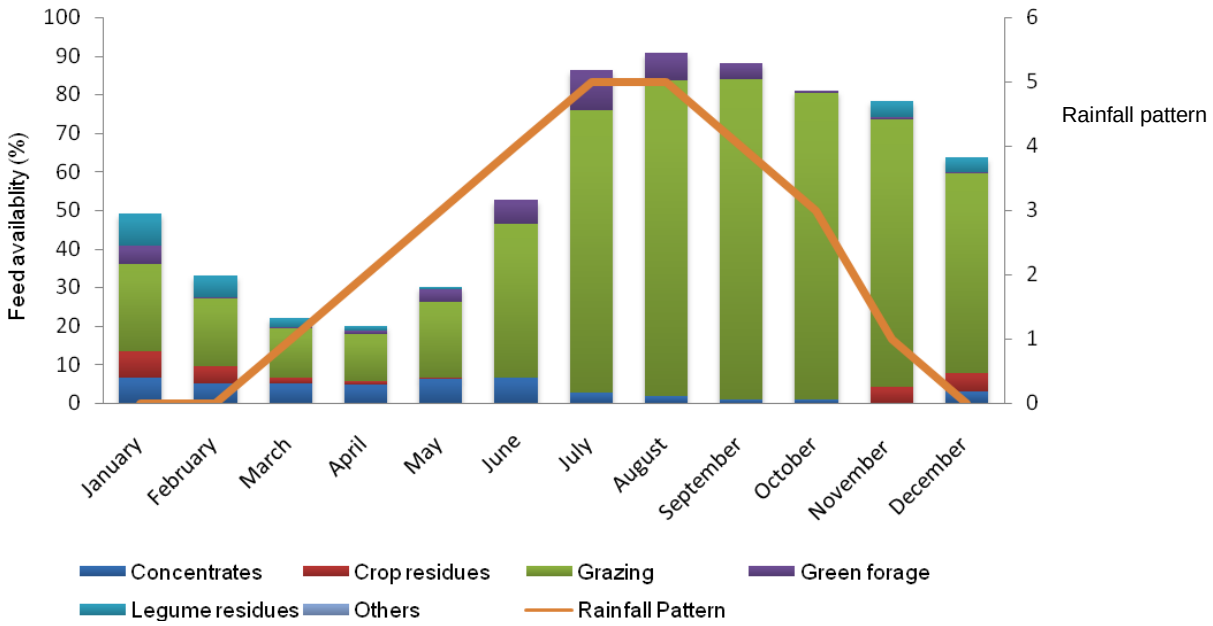

Figure 9. Available feed resources in Mahon. Rainfall pattern is on a scale of 0 to 5, where 5 = heavy rainfall and 0 = no rainfall.

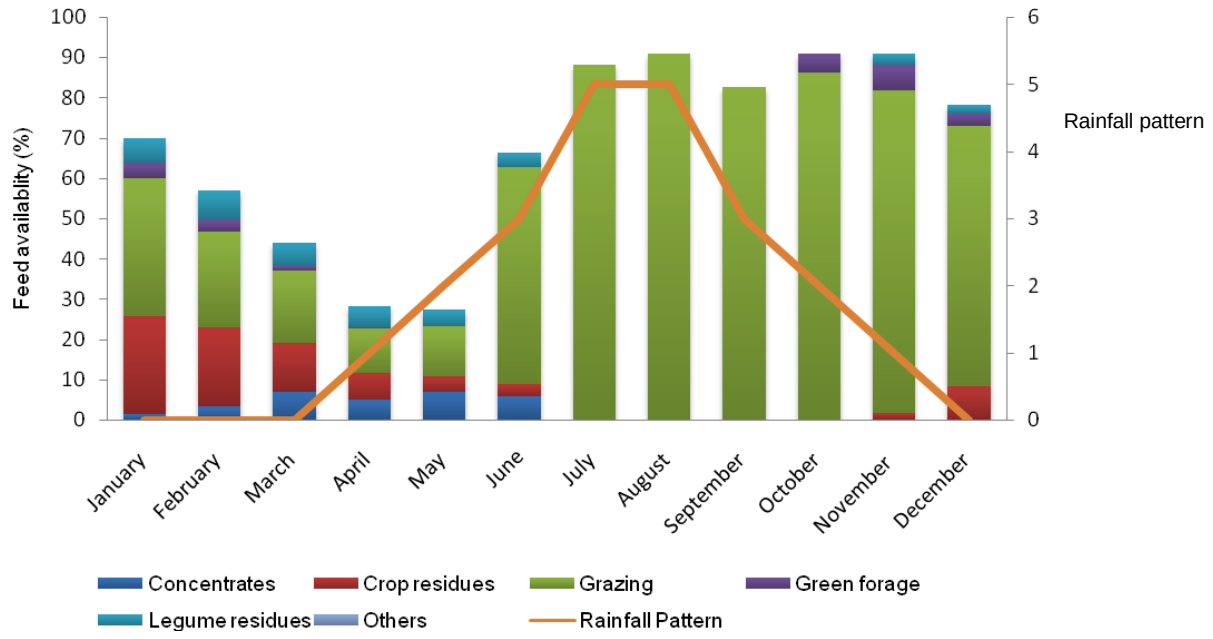

Figure 10. Available feed resources in Sayaga. Rainfall pattern is on a scale of 0-5, where 5 = heavy rainfall and 0.

wastes from its processing for making the local condiment. The pulp was mixed with chopped straw and feed to animals. This feed resource was the cheapest as at the time of the study and this may influence its choice and the quantity purchased. From the results, a total of 500 kg of *P. biglobosa* pulp was recorded as purchased by four of the respondents in Mahon at different intervals between May, 2013 and April, 2014. Maize bran and *P. biglobosa* (pulp) were commonly purchased in both sites, however, the quantity of each purchased feed was

Table 1. Quantity, prices and frequency of purchased feed in Mahon and Sayaga over the past 12 months.

Purchased feed	Number of farmers	Prices* (FCFA/kg)	Quantity purchased (kg)	Number of farmers	Prices* (FCFA/kg)	Quantity purchased (kg)
	Mahon			Sayaga		
Commercially mixed ration	6	220.00	375	4	200.00	1250
Cotton (*Gossypium* sp.) - seed cake	1	500.00	25	-	-	-
Cowpea – hay	1	100.00	25	-	-	-
Maize - bran	4	120.00	225	3	120.00	750
Parkia biglobosa (pulp)	4	50.00	500	6	50.00	2625

*Prices as at the time of the survey (500 FCFA = 1 USD).

higher in Sayaga than in Mahon.

Farmers in Sayaga depend more on livestock production than in Mahon as shown in the income generation from livestock. This might have influenced their decision for more purchased feed especially during feed shortage. There were slight variations in the prices of commercially mixed ration and *P. biglobosa* pulp in both study sites. Out of all the respondents in Mahon, 10% purchased cotton seed cake once in the last 12 months and no farmer in Sayaga purchased cotton seed cake which costs 500 FCFA per kilogram as at the time of the study. The low quantity and frequency of purchased feeds in both sites could be attributed to poor accessibility and the management practices in both Mahon and Sayaga. This suggests the need to improve the utilization of the available feed resources and to identify other potential feed resources and strengthening their use.

Feed quality

Grazing, including the natural pasture and crop residues left on the field, contributed the largest proportion to livestock diets in terms of DM, metabolizable energy (ME) and crude protein (CP) in the study sites followed by the naturally occurring green fodders which were collected for livestock feeding (Figures 11 and 12). While the percentage contribution of purchased feed to the DM, ME and CP in Mahon is less than 10%, purchased feed contributed 4, 18 and 8% of DM, ME and CP, respectively to the livestock total diet in Sayaga. This implies that farmers in Sayaga depend more on purchased feed than in Mahon. Trends in the contribution of collected crop residues to livestock feed were similar in both study sites. The percentage contribution of collected crop residues to the diets on the basis of DM, ME and CP was less than 3% in both study sites. It was noted that this percentage (3%) only accounted for the few collected quantity of crop residues. Farmers usually leave large percentage of crop residues on the farm for animal to graze which might have been mixed up or considered as grazing during the interview.

Generally, crop residues in both areas are mainly composed of cereals as the dominant crops. The feeding value of cereal stovers are very low while haulms of leguminous crop are very high owing to higher protein content (Carangal and Calub, 1987) but the quantities generated in these areas are generally low. This is owing to the fact that legumes are not commonly cultivated in the area. The quality of the collected crop residues become very low especially during the dry periods due to poor storage practices as residues are stockpiled on rafters outside where they are subjected to unfavorable weather and consequently, the quality diminishes as the residues lignify. Delayed harvesting can also lead to greater loss of leaves and leaf sheaths, the most digestible parts of cereal straws with a consequent reduction in nutritive value (Williams et al., 1997). Cultivated fodder, although not commonly practiced in Mahon, contributed about 1% of DM, ME and CP to the total diet of the existing feed resource. Although its contribution may be limited, it thus implies that cultivated pasture especially forage legume could improve the quality of livestock feed resources if integrated into arable cropping.

Farmers' knowledge of potential feed resources

In both study sites, farmers identified some potential feed resources and feed processing methods that they were aware of but are not currently practiced. They also mentioned few ways to strengthening the use of such resources (Table 2).

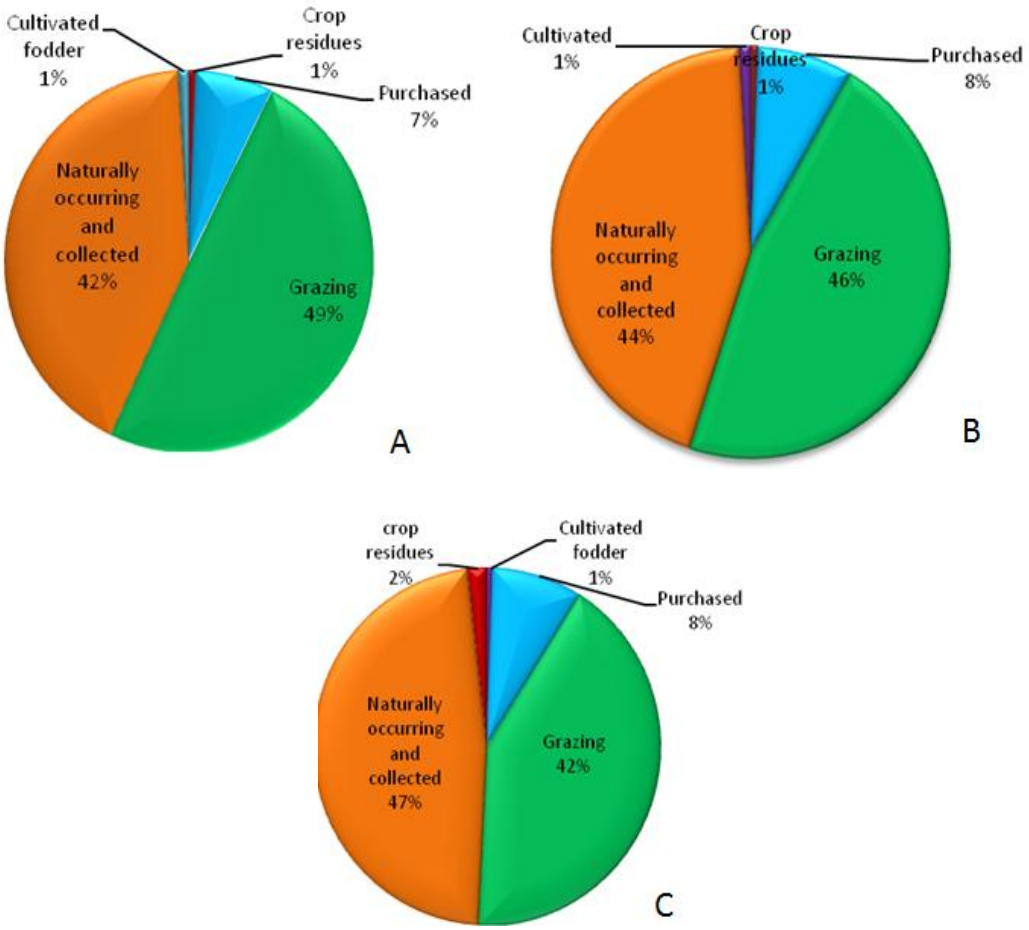

Figure 11. Contribution of various feed sources to the DM, ME and CP contents of total diet in Mahon respectively.

Composition of available feed resource during the dry season

In the dry season, available feed resources in both study sites ranged from herbaceous grasses (*Andropogon gayanus*) to browse trees, crop residues and crop by-products (maize bran, *P. biglobosa* pulp) (Tables 3 and 4). The species of browse trees found in the study areas were among the Sahelian and Sudanian browse species reported by Bognounou et al. (2008) and Sawadogo et al. (2010) as livestock feed. Schmidt et al. (2010) reported that the highest diversity of browse species was found in the Sudanian zone, where the vegetation consists of dry and sub-humid tree savannas and forests.

The study results indicated that a considerable number of browse species contributes to the ruminants' nutrition in the sub-Sahelian agro-ecological zone of Burkina Faso, particularly during the cool and hot dry season, which correspond to their fructification period (Heuzé and Tran, 2011). The mean value of crude protein (CP) concentration of the browse species available during the dry season in Mahon and Sayaga of 12.3 and 12.6% respectively doubled that of the crop residues and by-products of 6.12 and 4.12%, respectively. As discussed earlier, poor storage methods and leaving crop residues on the field, particularly for cereals reduced their feed values (Hiernaux et al., 2009). From the study results, the nutritional quality of legume residues and by-products need to be exploited as supplements particularly for lactating animals and animal fattening (Savadogo et al., 2000) by encouraging more intercropping with cereals. The use of browse as a source of protein could help livestock to cope with aggravated nutritional stress (Craine et al., 2010).

As the pastoralists and their livestock transited from the more arid Sahelian region in the north to the more humid Sudano-Guinean regions of the West Africa (Ayantunde et al., 2014), there is an anticipated increased pressure on forage resources in the region. In view of this, propagation and yield studies as well as fodder bank establishment and management should also be envisaged with other preferred browse species of high nutritional value that are drought tolerant and adapted to this regional. Species such as *Pterocarpus* which are

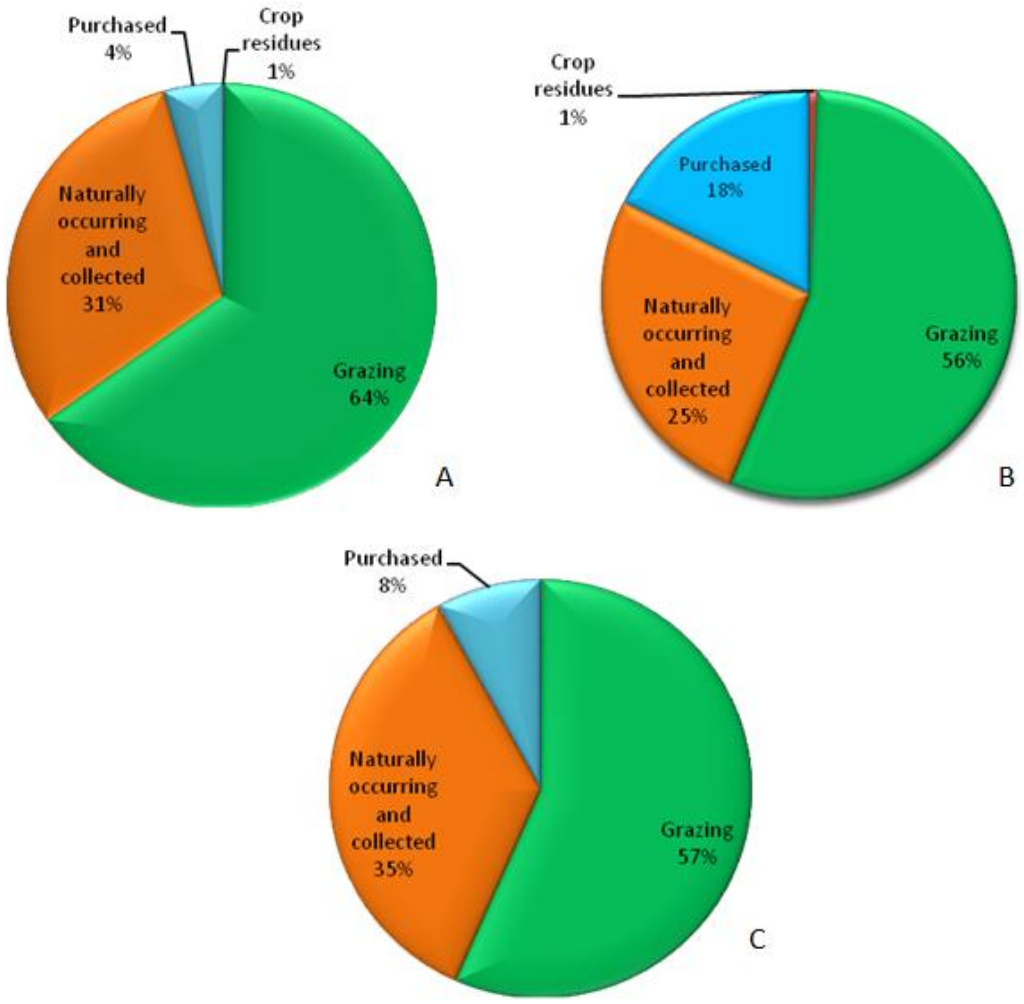

Figure 12. Contribution of various feed sources to the DM, ME and CP contents of total diet in Mahon respectively.

most frequently used by cattle, can be cultivated to yield a high amount of leaf biomass in Burkina Faso; they should therefore be introduced in local agroforestry systems to reduce dry season fodder scarcity (Ouédraogo-Koné, 2008).

Farmers' feed quality assessment indicators

In both Mahon and Sayaga, all the farmers interviewed confirmed the use of physical indicators to determine feed quality given to the animals (Table 5). All the farmers considered sick animals and pregnant female in that order as the priority animals that should be given quality feeds before other animals. Farmers reported that quality feeds are needed by sick animals for early recovery and for foetal development in pregnant females. Colouration, texture, odour, leaf to stem ratio, moistness, age at harvest and animal behaviour were listed as farmers'quality assessment indicators of livestock feed in both sites (Table 5).

In terms of colour as indicator, feeds generally have their own typical appearance, which the farmers are familiar with. Deviation from the typical appearance or colour was reported to have implication on quality. Farmers are able to assess the importance of feed quality as related to colour especially in the case of cereal residues. Berhanu et al. (2009) confirmed the use of visual observation and smell by farmers in Ethiopia to assess the quality of agro-industrial by-products as livestock feed and assessed black colouration of sorghum stover as poor quality and greenish colour hay good quality legume hay.

Another indicator for evaluation of feed quality reported by the farmers was texture. Soft and tender plant fraction and crop residues were considered high quality than the tough samples. For cereal stover, coarse texture of the stem residues with the presence of the sheath indicates

Table 2. Potential feed resource, feed processing methods and farmers' constraints in utilizing them.

Potential feed resources	Reason for not using the resource	How to strengthening the use of the feed resources
Mucuna pruiens	Knowledge gap is the major problem. Some of the farmers observed that when it was introduced to the village some years back, it's potential to add to feed quality and quantity was great. But they lacked the agronomic knowledge since it is an annual forage legume that should be planted yearly from the harvested seed.	Seed supply and knowledge of its annual propagation.
Leuceana leucocephala	Farmers reported lack of knowledge. One farmer watched it on the television once when he visited a relative in the city. Sharing with others, they are longing to know how to get access to the planting materials.	Supply of planting materials and training on how to be used properly.
Fodder maize and Forage grass	Lack of information and seed supply. The farmers remarked that they rarely see the extension service who will put them through and initiate any useful practice which they will then follow after.	Strengthening the capacity of the extension service. Adequate supply of the seed of fodder maize.
Methods of improving existing feed resources		
Conservation of crop residues	Lack of information and proper training on how to conserve the quality of crop residues.	Since maize is a major crop in the area and maize straw is of low nutritive value, convenient and affordable methods of improving maize straw quality should be sought.
Urea treatment of crop residues	Farmers reported that lack of knowledge was the predisposing factor for not exploiting the potential of urea treatment. The fear of losing the animal if the urea was not applied in the right quantity.	Awareness and training of farmers on the use of urea .

higher quality than smooth texture. Maize was ranked higher due to the presence of more sheaths which the farmers considered as an indication of quality as against sorghum and millet with lesser sheath. This confirmed a report that maize stover has a higher nutrient content than most straws (Suttie, 2000). Owen (1994) reported that animals are able to select for the more nutritious leaf and leaf-sheath components which contain more nitrogen against the less nutritious stem. As reported by the famers, coarse maize bran indicated high quality. Ground feeds with a large percentage of fine particles (<0.5 mm) are clearly less well accepted by cattle than coarse ground feeds (Morand-Fehr et al., 1994). The preference for coarse particles may be due to greater ease of prehension.

Leaf to stem ratio of forage crops is an important factor affecting diet selection, quality, and intake of forages (Smart et al., 2001), because leaves have usually higher nutrient quality (for example crude protein) than stems (Bakoglu et al., 1999). Farmers ranked higher in terms of quality the available sample of groundnut haulms with higher leaf to stem ratio than cowpea haulms. In an experiment, Savadogo (2000) reported groundnut haulms contained more leaves (48.1%) than cowpea haulms and both cowpea and groundnut haulms had been reported as high value supplement to poorer quality hay and stover to improve nutrient supply and growth of livestock (Savadogo et al., 2000; Bayala et al., 2014) with differing quality based on provenance, season and stages of maturity. This result underscores farmers' perception of quality based on leaf to stem ratio. Studies of the voluntary intake of sheep and cattle fed separated leaf and stem fractions of a range of tropical grasses have also indicated that leaf is eaten in much larger quantities than stem due to a shorter time the leaf is retained in the recticulo-rumen (Minson, 1980).

Farmers' perceived high moisture content relative to dryness as indicator for high feed quality as it reduces water intake by animals. Water intake (from all sources) had been reported to be related to the intake of dry matter (Forbes, 1985). The more moisture supplied in the feed, the lower the need for drinking water. Studies have also shown that the nutritive quality of forages varies as they grow towards maturity which agrees with farmers' assessment (Bilal et al., 2001). According to the farmers, animal behaviour is another indicator of feed quality assessment. Kalio et al. (2006) confirmed that the utilization of any feed resource is influenced by the animals' preference and acceptability of the feed, which is related to the animal's behavioral

Table 3. Nutrient composition (g/kg DM), metabolizable energy (MJ/kg) and *in vitro* organic matter digestibility (g/kg DM) of available feed resource in Mahon in dry season.

Type of feed	Ash	N	CP	NDF	ADF	ADL	ME	IVOMD
Grasses/Browse								
Khaya senegalensis (leaf)	127	24.0	149	425	34.7	74.2	71.7	522
Cassia sieberiana pods	47.8	12.9	80.5	325	31.6	107	90.9	610
Piliostigma tonenji pods	116	12.3	7.72	555	53.5	239	66.5	469
Cordia myxa (leaf)	141	24.1	151	595	67.2	279	55.4	423
Daniella oleifera (leaf)	63.7	30.3	18.9	569	42.2	137	102	705
Mangifera indica (leaf)	86.1	13.0	81.5	450	38.9	116	65.2	458
Cassia sieberiana (leaf)	42.9	21.2	133	665	55.4	249	71.5	501
Mean values	**89.2**	**19.7**	**123**	**512**	**46.2**	**172**	**74.7**	**527**
Crop residues/by-products								
Maize bran	11.9	11.7	72.9	389	8.62	13.6	119	776
Cowpea haulms	91.7	27.2	170	459	24.2	60.1	92.2	648
Groundnut haulms	239	18.7	117	452	44.7	152	72.6	539
Parkia biglobosa pulp	43.3	06.6	40.9	185	16.27	06.4	124	805
Millet stover	24.7	02.2	13.9	641	71.1	147	52.4	350
Maize stover	216	07.0	43.5	672	42.5	76.5	78.8	556
Maize husk	39.3	03.0	18.8	871	43.1	49.8	88.6	579
Sorghum bicolor stover	43.8	02.2	14.0	861	64.8	120	55.7	375
Mean values	**88.7**	**0.98**	**61.2**	**566**	**39.4**	**78.1**	**85.4**	**579**

Nitrogen (N), crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF), acid detergent lignin (ADL), Metabolizable energy (ME) *in vitro* organic matter digestibility (IVOMD).

pattern. Similarly, the extent of preference and acceptability of a feed by the animals is one of the principal things to note when determining the best feed stuff (Ganskopp et al., 1997). The result suggests that there is significant complementarity between farmers' assessments of feed value of the available feed resources and relative assessment derived from laboratory analysis (Thorne et al., 1999).

Constraints to livestock production and proposed solutions

The livestock production constraints were identified in order of importance and farmers suggested solutions (Table 6). Shortage of water was described by the farmers as the first constraint in both communities. Inadequate quantity and quality feed throughout the year ranked next to shortage of water.

Opportunities for efficient conservation and utilization of available feed resources

The followings are potential interventions for both Mahon and Sayaga derived from farmers' proposed solutions and existing opportunities:

1. Farmers reported that lack of information on other feed resources was the predisposing factor for not exploiting the potential of the livestock sector to the fullest. Hence, training on ration formulation based on the locally available feed resources, fodder production, feed conservation and processing would be instrumental to enhance knowledge base and attitude/behaviour change of the farmers and livestock extension workers.
2. Improving the utilization of crop residue
a. Large quantities of crop residues are produced from cereals. However, only few are collected and properly used as animal feeds throughout the season due to poor storage facilities and strategies. Conservation of crop residues should be encouraged. Farmers need to be trained on how to conserve crop residues in a rain proof, well-ventilated barn to retain the quality of the crop residues throughout the period of storage.
b. Crop residue treatment and crop residue-based rations. In both villages, cattle showed symptoms of allotrophagia, a condition arising from mineral deficiency. On-farm training on ration formulation using cereals and legumes crop residues will increase the quality of livestock feed in the areas. Farmers could also be trained on the making of multinutrient feed block using local resources to improve animal feed intake.
3. Land was reported as one of the limiting factors inhibiting expansion of agricultural activities in the areas.

Table 4. Nutrient composition (g/kg DM), metabolizable energy (MJ/kg) and *in vitro* organic matter digestibility (g/kg DM) of available feed resource in Sayaga in dry season.

Type of feed	Ash	N	CP	NDF	ADF	ADL	ME	IVOMD
Grasses/Browse								
Andropogon gayanus (leaf)	147	05.7	35.7	743	529	109	53.5	385
Daniella oliveri (leaf)	69.9	31.6	198	609	475	189	67.7	498
Prosopis africana (leaf)	93.5	14.9	92.9	833	417	225	42.9	323
Pterocarpus microcarpa (leaf)	101	36.3	227	451	376	129	79.6	585
Vitellaria paradoxa (leaf)	48.7	15.1	94.1	506	469	198	50.9	365
Tectonia grandis (leaf)	86.2	40.5	253	421	338	141	72.2	542
Piliostigma tonenji pods	84.1	12.0	75.2	569	540	223	62.7	439
Dicrostachys cinera (leaf)	47.4	29.5	184	485	424	223	54.1	406
Cordia Africana (leaf)	99.5	17.4	109	503	422	198	61.0	440
Flugia virosa (leaf)	69.2	27.9	174	309	253	71.7	60.8	449
Cordia africana pods	135	16.4	102	395	404	178	45.4	348
Trichilia ematica (leaf)	69.5	17.8	111	502	421	128	93.1	635
Mean values	**79.4**	**20.2**	**126**	**516**	**422**	**149**	**76.1**	**535**
Crop residues/by-products								
Sorghum bicolor stover	80.0	03.8	23.5	654	437	60.8	75.0	503
Millet stover	106	02.5	15.6	721	524	83.4	74.0	499
Maize bran	146	13.5	844	351	99.0	16.9	78.6	552
Mean values	**111**	**06.6**	**4.12**	**57.5**	**354**	**53.7**	**75.9**	**518**

Nitrogen (N), crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF), acid detergent lignin (ADL), Metabolizable energy (ME), *in vitro* organic matter digestibility (IVOMD).

Table 5. Farmers' indicators of feed quality assessment.

Parameters	Description
Colouration	Dark green colouration showed better quality than yellowish green. For cereal crop residues, no specific colour was mentioned, however, farmers noted that green colouration of cowpea and groundnut crop residues indicated good quality.
Texture	Farmers reported that coarse texture of maize gluten with bran due to the presence of crushed grains are of higher quality that the powdery maize gluten with bran. Coarse texture of the stem sorghum residues with the presence of the sheath indicated higher quality than smooth texture. Soft and tender plant fraction and crop residues were considered high quality than the tough samples.
Odour	Farmers described an offensive odour (pungent and rancid smell) in commercially formulated ration as poor quality while a pleasant smell of fresh coconut indicated high quality.
Leaf to stem ratio	Farmers considered high leaf to stem ratio as high quality feed for both fresh pasture and crop residues.
Moistness	From farmers' perspective, high moisture content indicated high quality. Some farmer reported that when animals take moist feed, their water intake will be reduced and this help to them manage the limited water source available because water supply was a major constraint.
Age at harvest	Farmers reported that younger plants are of higher quality than older plants.
Animal behaviour	Animals exhibited certain behaviour that indicated the quality of a feed. Wagging of tails when the food is served, competition for the feed, high intake rate and no left over are indicators of high quality as described by the farmers.

Therefore, planting of forage legume trees as fodder bank around the homestead and backyard as forage production is recommended. The legume trees are multipurpose and can provide feed, fuel, serve as live fences and shade. Backyard production of fodder trees is not land or labour demanding.

Table 6. Paired wise matrix ranking of major problems identified by the farmers facing livestock production in Mahon and Sayaga and suggested solutions.

Problems in order of importance	Problems identified	Proposed solution by the farmers
1	Shortage of water in the dry season for animals	- Construction of small dam for the animal. - Provision of water storage facilities in the village.
2	Inadequate quantity and quality feed throughout the year.	- Training and skill acquisition on how to improve the quality available feeds especially crop residues - Training on how to formulate animal feed ration using available feed resources. - Provision of forage legume and grass seed for cultivation - Training on proper storage methods for crop residues.
3	High cost of veterinary drugs and services	Establishment of a veterinary service and drug centre in the village.
4	Poor performance of their local breeds	Introduction of improved breed for cross breeding programme that can improve the genetic capacity of the local breed.
5	Poor housing provision	Provision of concrete, ventilated and protective housing for the animals. This will also facilitate the collection and utilization of animal dungs as manure.

a. Integrated crop farming with shade-tolerant forage grass and legumes planted in tree plantation. This practice encourages the effective utilization of same land resources where the tree biomass helps to enrich the soil and the animal dungs as well. Planting of fodder shrubs and tree as edge rows in arable crop land in form of alley-cropping will also restores nitrogen to the top layer of soil so that farmers can use the same piece of land year after year to grow their forage crops.
b. General health management and diseases prevention are the most important factors that affect ruminant production in the tropical environments. Losses because of ill health and diseases have not been quantified in the economic terms. Therefore, intervention must include proper animal healthcare. Provision of a veterinary center around these two villages is required.
c. Better management of the existing water resources, collecting rain water for dry periods, utilizing the existing limited water sources by making water reservoirs or ponds. Extracting the ground water with the assistance of government and non-government organizations.

CONCLUSION AND RECOMMENDATIONS

The farming system in both Mahon and Sayaga was characterized by mixed crop-livestock production system with fruit trees as major crops. Majority of the farmers are smallholder cultivating less than 5 has of land. Cattle are the most important livestock in these areas and are predominantly local breeds. Agricultural practices in both areas were characterized majorly by fruit trees cultivation and then arable crops. This contributes significantly to household's income generation. Livestock rearing contributes next to crop farming. Livestock production in these areas depends mostly on grazing of natural pastures. Crop residues, though available, had less contribution to the livestock diet in terms of DM, ME and CP, particularly when critically needed between March and April. This is as a result of poor storage facilities and continuous cropping on same land which affect the quality of the crop and residue. Although inadequate quantity and quality feed throughout the year was a problem facing livestock production in both Mahon and Sayaga, farmers viewed that shortage of water in the dry season for animals was the main constraint to livestock production in the areas. It is important to address the improvement of utilization of crop residues since it is available in both study sites. It is also important to encourage forage production to improve livestock productivity and farmers' livelihood.

Conflict of Interests

The authors have not declared any conflict of interests.

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Full Length Research Paper

Applications of magnetic technology in Agriculture as a novel tool for improving crop productivity (1): Canola

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Two field trials using canola (var. Serw-6) were conducted to study and evaluate the effects of magnetizing irrigation water on Canola vegetative growth, photosynthetic pigments, seed, yield and water use efficiency as well as seed biochemical constituents and fatty acids composition of the yielded oils. Application of magnetizing irrigation water led to marked increases in growth parameters (plant height (cm), fresh weight and dry weight (g plant^{-1}) and water contents (%); photosynthetic pigments (chlorophyll a, chlorophyll b, chlorophyll (a+b), carotenoids and consequently total pigments). Treating plants with magnetized water increased also, seed yield and its components plant height (cm), branches (number plant^{-1}),seed weight (g plant^{-1}), pods (number plant^{-1}), seeds weight (g pod^{-1}), 100-seed weight (g) and seed yield (kg fed^{-1}) as well as seed biochemical constituents (oil (%), oil yield (kg fed^{-1}) macro and micro elements). The treatment improved oil quality as it affected fatty acids composition of canola oil, by increasing total unsaturated fatty acids and total essential fatty acids. Moreover, Water Use Efficiency (WUE) increased significantly as a result of irrigation with magnetic water by 19.05% compared to control plant. The present findings have shown that irrigation with magnetized water could be employed as one of the most valuable modern technologies that can assist in saving irrigation water and improving yield and quality of Canola under newly reclaimed sandy soil. The usage of magnetic water in the agricultural production will enable intense and more quantities and qualitative production.

Key words: Canola, magnetic water, water-use efficiency, nutritive value, oil, fatty acids.

INTRODUCTION

Water is an unusual substance, mostly due to its 3D network of hydrogen bond in the molecule. Its properties allow it to act as a solvent, as a reactant, as a molecule with a cohesive properties, as an environment and a temperature stabilizer (Ibrahim, 2006). No other liquid can replace water. It has relatively high melting and boiling points for a small molecule, high specific heat capacity, and higher density of liquid than that of solid

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(Eisnberg and Kauzmann, 1969). Several investigations into electromagnetic effects on plants have been carried out with some remarkable results. The optimal external electromagnetic field could accelerate the activation of seed germination (Maeda, 1993; Oomori, 1992), however, the mechanism of these actions is still poorly understood (Morar et al., 1988; Xiyao et al., 1988). Electric and/or magnetic treatments are assumed to enhance seed vigour by influencing the biochemical processes that involve free radicals and by stimulating the activity of proteins and enzymes (Kurinobu and Okazaki, 1995).

Magnetic field (MF) became a part of the environment and source of energy, thereby effects normal metabolisms (Belyavskaya et al., 1992) and has impact on meristem cell division (Aladjadjiyan, 2007). In addition, MF affects water absorption, preservation and ionization (Taia et al., 2007). Forces generated by MF may cause magnetophoresis in macromolecules (Paul et al., 2006). Metabolic substances as plants photosynthetic pigments could be affected by MF. It has been found that an increase occurs in chemical reactions of plants under MF, which has a positive effect on photochemical activity, respiration ratio and enzyme activity (Phirke et al., 1996; Martinez et al., 2000; Carbonell et al., 2000).

Oilseed plant has become a plant of major agro-economic importance, with a seed yield of 47 millions tones worldwide in 2007 (FAO, 2007). It is considered as one of the three major oil crops in many countries especially Canada, European Union and USA because it has a wide range of uses (oil production, animal feeding, alternative fuel, etc) (Howlett et al., 2001; Abdallah et al., 2010). Cultivation of canola in Egypt may provide an opportunity to overcome some of the local deficit of vegetable edible oil production, particularly it could be successfully grown during winter season in newly reclaimed land outside the old one of Nile valley to get-around the competition with other crops occupied the old cultivated area (Kandil, 1984; Sharaan, 1986; Ghallab and Sharaan, 2002; Sharaan et al., 2002; Megawer and Mahfouz, 2010). Suitability of growing canola under Egyptian conditions, compared with other oil crops, may be ascribed to its tolerance to harsh environmental influences frequently prevailing in such newly reclaimed soil such as salinity and drought (Weiss, 1983).

The target of this work is to increase the efficiency of growth and productivity of canola plant grown under environmental stresses by using magnetized water.

MATERIALS AND METHODS

Two field trials using canola (var. Serw-6) were conducted at Research and Production Station, National Research Centre, Alemam Malek Village, Al-Nubaria District, Al-Behaira Governorate, Egypt in 2009/10 and 2010/11 winter seasons to study and evaluate the effects of magnetizing irrigation water on growth, photosynthetic pigments, yield and yield components of canola winter crop. The experimental soil and water were analyzed according to the method described by Chapman and Pratt (1978) (Table 1).

Cultivation method and layout of experiment

The soil of experiment was ploughed twice and divided into plots (10 length m x 5 m width). Recommended rates of canola seeds (3 kg/fed; variety Serw-6; fed=4200 m^2) were sown by drilling seed manually in the rows at 15-cm apart at the first week of November in both seasons. Four replications were used in each treatment. Control treatment was irrigated with normal water, while the other treatment (magnetized water) was irrigated with water after magnetization through a two inch Magnetron [U.T.3, Magnetic Technologies LLC PO Box 27559, Dubai, UAE]. Phosphorus and potassium fertilizer were added before sowing at the rate of 200 kg/fed. as super phosphate (15.5 % P_2O_5) and50 kg/fed potassium sulphate (48 to 50% K_2O), respectively, while nitrogen fertilizer was added at the rate of 45 kg N/fed as ammonium nitrate (33.5%N) in two equal doses at 21 and 35 days after planting (DAP), respectively. Sprinkler irrigation was applied as plants needed. The layout of experiment was shown in (Figure 1).

Data recorded

Growth parameters

After 85 days from sowing, 10 plants from each treatment were cutting on 5 cm above ground to determined vegetative growth, that is, plant height, fresh and oven dry weight. Water content was determined according to (Henson et al., 1981) using the following formula:

WC = 100×(fresh mass – dry mass)/fresh mass.

Yield and yield components

At harvest time (180 day after sowing), a random sample of 20 plants from each plot were taken to determine some yield attributes such as number of siliqua/plant; number of seeds/siliqua, seed yield/plant (g) and 1000-seed weight (g). The whole plot was manually harvested to determine the above ground biomass (biological yield) after dried under sunshine for one week, pods were threshed to determine seed yield; straw yield was calculated by subtracting seed yield from biological yield; harvest and crop indexes were calculated by dividing seed yield/biological yield and straw yield, respectively.

Water-use efficiency (WUE)

WUE values were calculated with the following equations (Howell et al., 1990).

$$\mathrm{WUE} = (\frac{E_y}{E_t}) \times 100$$

Where WUE is the water use efficiency (kg/m^3); E_y is the economical yield (kg/fed./season); E_t is the total applied of irrigation water, m^3/fed/season.

Photosynthetic pigments

Total chlorophyll a and b and carotenoids contents in fresh leaves were estimated using the method of (Lichtenthaler and Buschmann,

Table 1. Soil and water analysis for site experiments.

Parameters	Soil depth (cm)		Irrigation water	
	0-15	15-30	Before magnetic	After magnetic
Particle size distribution				
Coarse sand	48.20	54.75	..	…
Fine sand	49.11	41.43	..	…
Clay + Silt	2.69	3.82	..	…
Texture	Sandy	Sandy	..	…
pH (1:2.5)	8.22	7.94	7.25	7.13
EC (dSm^{-1})(1:5)	0.20	0.15	0.50	0.40
Organic matter (%)	0.67	0.43	…	
Soluble cations (mq/l)				
Ca^{++}	0.60	0.50	2.15	2.05
Mg^{++}	0.50	0.30	0.50	0.65
Na^{+}	0.90	0.80	3.00	3.00
K^{+}	0.20	0.10	0.31	0.31
Soluble anions (mq/l)				
CO^{-3}	-	-	0.01	0.01
HCO^{-3}	0.60	0.40	2.33	2.46
Cl^{-}	0.75	0.70	2.17	1.72
SO^{-4}	0.85	0.60	1.45	1.82

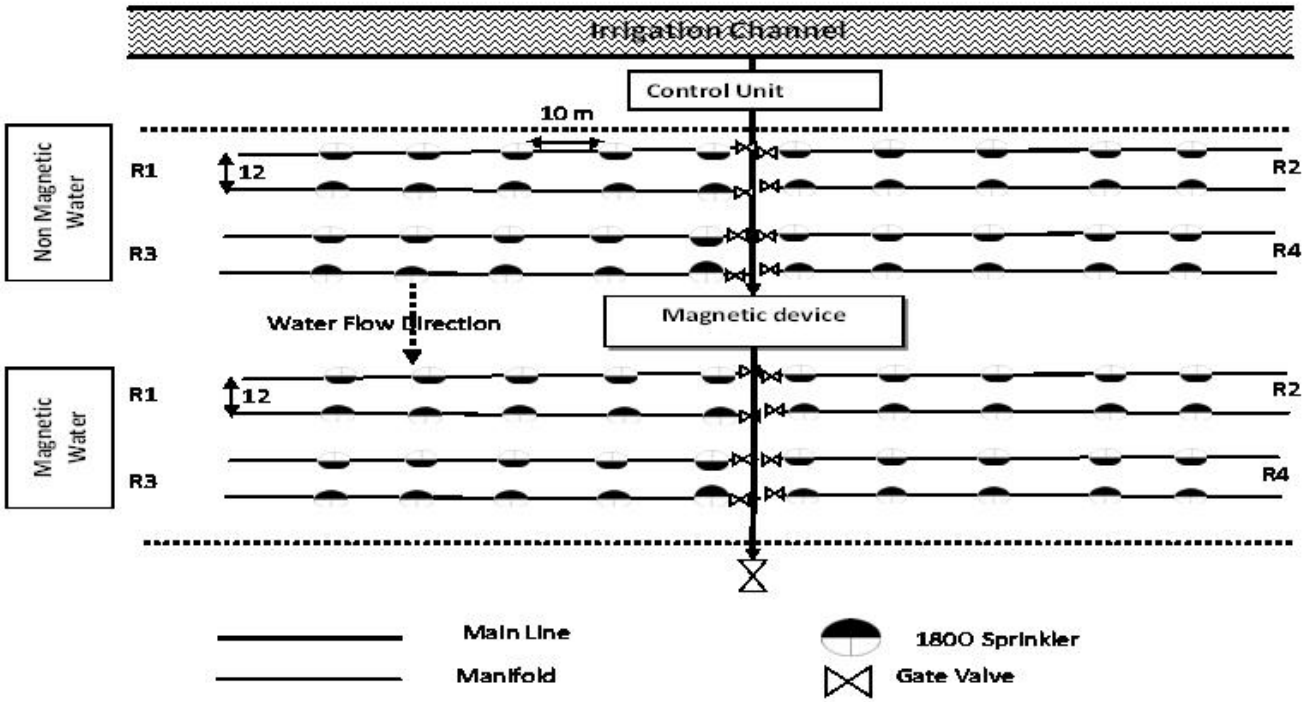

Figure 1. Layout of experiment design under solid set sprinkler system.

2001). The fresh tissue was fine ground in a mortar and pestles using 80% acetone. The optical density (OD) of the solution was recorded at 662 and 645 nm (for chlorophyll a and b, respectively) and 470 nm (for carotenoids) using a spectrophotometer (Shimadzu

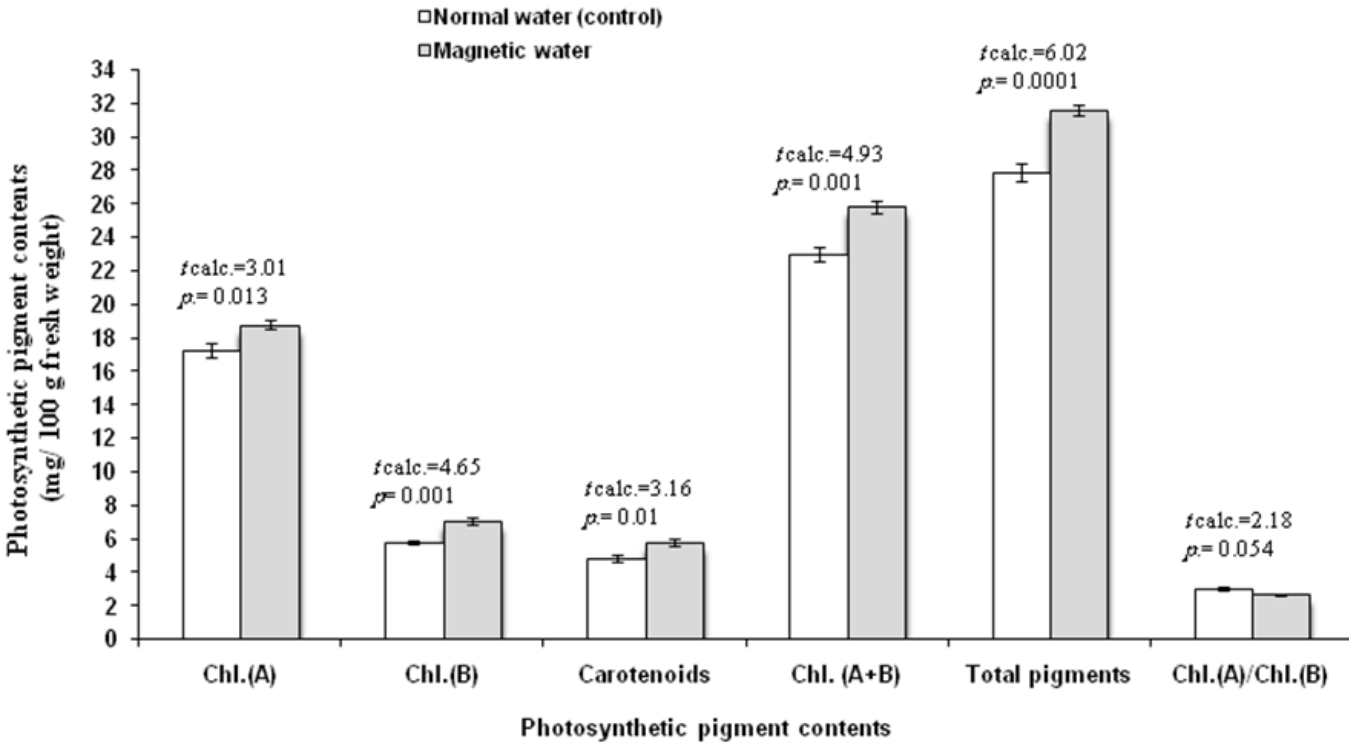

Figure 2. Effect of irrigation treatments on photosynthetic pigment contents (data average of 2009/10 and 2010/11 seasons). N=6, *, **, *** *t* is significant at the *P* **<** 0.05, 0.01 and 0.001 levels, respectively, P-value **>** 0.05 non-significant.

UV-1700, Tokyo, Japan). The values of photosynthetic pigments were expressed in mg/100 g FW. At harvest yield, yield components and quality of Canola crop were determined.

Oil determination

The oil of Canola seeds were extracted according to Kates and Eberhardt (1957). The powdered seeds were shaken overnight with isopropanol: chloroform (1:1). The solvent were evaporated under reduced pressure of CO_2 atmosphere. The lipid residue is taken up in a chloroform: methanol (2:1 v/v) and given a folch wash, the dissolved total oils were purified by washing with 1% aqueous saline solution. The aqueous phases were washed with chloroform that was combined with the pure oil solution. Chloroform was evaporated and the total pure oil was weighed.

Fatty acid determination

To oil sample 20 ml methanol, 10 ml benzene and 1ml concentrated sulphoric acid were added in glass tube and refluxed for 90 min, the methyl esters obtained were extracted with petroleum ether (b.p. 40 TO 60ºC). The petroleum ether was then evaporated; the residue was dissolved in chloroform (Harborne, 1984). The methylated samples were subjected to analysis by Gas Liquid Chromatography (GLC) equipped with dual flame ionization detector and dual channel recorder.

Nutritional value of yielded seeds

The dried seeds were fine ground to determine K, Mg, Ca, Na, Fe, Mn and Zn concentration as described by Cottenie et al. (1982).

Statistical analysis

Statistical analysis was carried out using SPSS program Version 16 (SPSS Inc., 2005). Independent *t*-test was also carried out to find the significant differences between magnetic and nonmagnetic water treatments.

RESULTS AND DISCUSSION

Photosynthetic pigment contents: Data in Figure 2 illustrate the photosynthetic pigment responses of canola plant irrigation with magnetized and normal water. The current study shows that photosynthetic pigments are significantly affected by the magnetic water where chlorophyll a, chlorophyll b, carotenoids and total pigments concentration recorded more value under magnetic water compared with plants irrigated with normal water (Figure 2). The percent of increments reached to 8.86, 22.22 19.71 and 1350% in the above parameters, respectively**.** Photosynthetic pigments are considered good criteria to monitor, explain and correlate the changes induced by stress, interacted with other treatments; it controlled the economic yield whether in direct or indirect manner especially under stress conditions. This result are in a good harmony with several

Table 2. Effect of irrigation treatments on growth parametrs and water content of Canola at 85 days after sowing (data average of 2009/10 and 2010/11 seasons).

Treatment / Character	Mean ± SE		*p-value*	Increase (+) (%) over control
	Normal water (control)	Magnetic water		
Plant height (cm)	78.80 ± 1.40	92.00 ± 1.81	0.001	16.75
Fresh weight (g plant^{-1})	65.26 ± 0.96	81.26 ± 1.44	0.001	24.52
Dry weight (g plant^{-1})	7.44 ± 0.13	8.11 ± 0.15	0.002	9.01
Water contents (%)	88.55 ± 0.27	89.96 ± 0.27	0.001	1.59

N = 15, **, *** *t* is Significant at the *P* < 0.01 and *P* < 0.001 level, respectively.

studies for different plants; where MF treatment increased the chlorophyll content in sugar beet (*Beta vulgaris* L.) leaves (Rochalska, 2005; Hozayn et al., 2013a) and content of chlorophyll a, b and carotenoids in potato (*Solanum tuberosum* L.) (Rakosy-Tican et al., 2005; Atak et al., 2003, 2007), found an increase in chlorophyll content appeared after exposure to a magnetic field for a short time. The stimulating effect of magnetic treatments on photosynthetic pigments may be due to increasing proline content, which increased some ions as Mg^{2+} needed for chlorophyll synthesis (Shaddad, 1990) and/or K^{+}, which led to increased photosynthetic efficiency possibly by increasing the number of chloroplasts per cell (Garcia-Reina and Arza, 2001). Also, the increase in the concentration of chlorophyll pigments due the magnetic treatments may be attributed to the increase in GA_3 content in plants (Selim et al., 2009), which led to increase in the green pigments in the treated plants by stimulating the production of chlorophyll in leaves (Bethke and Drew, 1992; Wafaa et al., 2007; Amira et al., 2010a, b; Hozayn et al., 2011) reported that, magnetic treatment increased photosynthetic pigment contents *via,* increasing growth promoters (IAA).

Growth parameters

Data in Table 2 show that irrigation canola plant with magnetized water caused significant increases in all growth tested parameters (plant height, fresh and dry weight of plant) and Relative Water Contents (RWC) of plant compared with plant irrigated with normal water. Data indicate that there was a significant increase in plant height, fresh and dry weights and water content, by 16.75, 24.52, 9.01 and 1.59%, respectively as compared with non magnetized water application. These results are in conformity with those obtained by (De Souza et al., 2006) on tomatoes, who found a significant increase in dry weights of root, shoot and whole plants as a result of treating plant with magnetic water. Flórez et al. (2007) observed an increase in the initial growth stages and an early sprouting of maize and rice seeds exposed to 125 and 250 mT stationary magnetic field. Martı´nez-Te´llez et al. (2002) observed similar effects on wheat and barley seeds magnetically treated. The mechanisms are not well known yet, but several theories have been proposed, including biochemical changes or altered enzyme activities by Phirke et al. (1996).

Yield and yield components

Table 3 show that yield and its components such as plant height (cm), branches (number plant^{-1}) , seed weight (g plant $^{-1}$), pods (number plant^{-1}), seed weight (g pod^{-1}), 100-seed weight (g), oil (%), seed yield (kg fed^{-1}) and oil yield (kgfed^{-1}) were significantly enhanced under irrigation with magnetic water. These results confirmed previous studies on wheat, flax, lentil, chickpea and sugar beet where magnetic treatment gave higher value of yield and yield components compared to control treatment (Hozayn and Abd El-Qodos, 2010a, b; Abd El-Qodos and Hozayn, 2010a, b; Hozayn et al., 2013a, b). Similar effects have been reported abroad on buckwheat, sunflower, flax, pea, wheat, pepper, tomato, soybean, potato and sugar beet yields (Gubbels 1982; Pietruszewski 1999; Takac et al 2002; Crnobarac et al 2002; Marinkovic et al., 2002). Regarding the increment in oil (%) and oil yield (kg fed^{-1}) with magnetic water, these increases might be due to the increase in vegetative growth and nutrients uptake. These results are in good agreement with those (Crnobarac, et al. 2002) showed an increase in yield of soybean from 5 to 25%, with a higher quantity of oil from 13.2 to 17.3%.

Water use efficiency

Water use efficiency (WUE) values were increased by the irrigation with magnetic water. Data recorded in Table (4) show that WUE for dry matter production of Canola was significantly increased as the result of application of magnetized water. These results are in good harmony with those obtained by Selim and El-Nady (2011). Water absorption by lettuce seeds previously treated in stationary magnetic field and found significance increase in the rate of water absorption accompanied with an increase in the total mass (Garcia-Reina and Arza, 2001).

Table 3. Effect of irrigation treatments on Canola yield and its components (Data average of 2009/10 and 2010/11 seasons).

Treatment / Character	Mean ± SE		*p-value*	Increase (+) or decrease (-) (%) over control
	Normal water (control)	Magnetic water		
Plant height (cm)	133.22 ±2.40	150.00 ± 2.57	0.001	12.60
Branches (number plant^{-1})	6.94 ± 0.37	7.93 ± 0.27	0.038	14.27
Seed weight (g plant $^{-1}$)	64.00 ± 1.69	96.80± 4.46	0.001	51.25
Pods (number plant^{-1})	11.82 ± 0.24	14.81 ± 0.41	0.001	25.30
Seeds weight (g pod^{-1})	0.186 ± 0.004	0.159 ± 0.010	0.029	-14.52
1000-seed weight (g)	4.23 ± 0.02	4.00 ± 0.03	0.001	-5.44
Oil (%)	28.00 ± 0.15	32.00 ± 0.20	0.001	14.29
Seed yield (kg fed^{-1})	502.53 ±12.83	697.00 ± 11.80	0.001	38.70
Oil yield (kg fed^{-1})	140.71 ± 2.68	223.04 ± 3.40	0.001	58.51

N = 15 for all parameters except seed and oil yield where N=8, *, **, *** *t* is significant at the *P* **<** 0.05, 0.01and 0.001 levels, respectively.

Table 4. Effect of irrigation treatments on water use efficiency (WUE) of Canola (Data average of 2009/10 and 2010/11 seasons).

Treatment / Character	Mean ± SE		*p-value*	Increase (+) or decrease (-) (%) over control
	Normal water (control)	Magnetic water		
WUE (kg seed m^3 water)	16.63	23.07	0.001	38.70
WUE (kg oil m^3 water)	4.66	7.38	0.001	58.51

N=8, *** *t* is significant at the *P* **<** 0.001 levels.

These beneficial effects of magnetic field may be due to the increase in ions up take (Duarte et al., 1997; Esitken and Turan, 2004), especially Ca^{2+}. In most studies in recent years, exogenous Ca^{2+} can enhance plant drought resistance, inhibit the synthesis of activating oxides, protect the structure of cellular plasma membranes and maintain normal photosynthesis as well as regulate the metabolism of plant hormones and other important chemicals (Song et al., 2008; Blum, 1993).

Fatty acid composition

Data presented in Table 5 show fatty acid constituents of Canola plants irrigated with magnetic and normal water. These fatty acids are palmitic (C16:0), Stearic (C18:0), Oleic (C18:1), Linoleic (C18:2), Linolenic (C18:3), and Beheric (C 22:0). However, the predominant saturated fatty acids were palmitic acid and stearic acid in the Canola plants, while Linoleic and Oleic acid were the predominant as unsaturated fatty acid. Concerning magnetic water treatment effect, it was evident that palmitic and stearic acids increased with application of magnetic water treatment when compared with that of the control treatment. Irrigation with magnetic water induced marked increases in the levels of unsaturated fatty acids particularly oleic acids. The magnitude of such increase was much more pronounced by applying magnetic water than that of normal water. Abdel Rahim et al. (2000) reported that the percentage of unsaturated fatty acids proved the efficiency of de-saturation in oil. There was also great increase in unsaturated fatty acid with slight increasing of saturated fatty acids and consequently, increasing in Tus/Ts. Thus the yielded oil becomes safer for human consumption.

Macro and micro elements

Table 6 presents the influence of magnetized water on micro and macronutrients of Canola plant. Magnetic water caused significant decreases in nitrogen by about 17.3% compared with non magnetized water as well as increased potassium, magnesium and calcium copper percent by about 3.5, 6.7 and 0.8% of Canola plant respectively (Table 6). Regarding the effect of magnetic water treatment on P percent, the results recorded non significant variation between treatments as compared with the corresponding control. With regard to the effect of MW on microelement contents of Canola, data (Table 6) revealed that, using magnetized water caused gradual decrease in Fe and Zn contents by about 7 and 18% respectively. In the meantime, magnetic water caused significant increase in both Mn and Cu microelement as compared with the corresponding control. Duarte et al. (1997) reported an increase in nutrient uptake by magnetic treatment in tomatoes. A marked increase in P content of citrus leaves by magnetically treated water

Table 5. Fatty acid composition seeds of Canola plants irrigated with magnetic and normal water.

Composition	Control	Magnetic water
Palmitic (C16:0)	3.01	3.62
Stearic (C18:0)	19.72	22.66
Oleic (C18:1)	13.57	34.10
Linoleic (C18:2)	9.71	-
Linolenic (C18:3)	-	-
Behenic (C22:0)	1.51	1.07
Total saturated (TS)	24.21	27.26
Total unsaturated (TUS)	23.28	34.10
TUS/TS	0.96	1.25

Table 6. Macro and micro elements in seeds of canola plants irrigated with magnetic and normal water.

Treatment / Character		Mean		*p-value*	Increase (+) or decrease (-) (%) over control
		Normal water (control)	Magnetic water		
Macronutrients (%)	**N**	2.49	2.06	0.04	-17.27
	P	1.30	1.30	ns	0.00
	K	0.90	0.93	ns	3.33
	Mg	0.30	0.32	ns	6.67
	Ca	1.35	1.36		0.74
Micronutrients (ppm)	**Fe**	121.50	113.40	0.001	-6.67
	Mn	55.00	60.00	0.030	9.09
	Zn	76.50	63.00	0.020	-17.65
	Cu	10.50	13.50	0.040	28.57

N=6, *, **, *** *t* is significant at the *P* **<** 0.05, 0.01and 0.001 levels, respectively, P-value **>** 0.05 non-significant.

was also reported by Hilal et al. (2002). Algozari and Yao (2006) reported that the magnetic application led to easy breakthrough of water for the cell membrane of plants. The easy breakthrough of water leads to better absorption of water and mineral by plant roots (Barefoot and Reich, 1992). Kronenberg (2005) showed that the magnetic application led to an increase in the availability of minerals in soil through increasing of solubility of salts and minerals. Increasing of solubility of salts and minerals led to the increasing of macro and micro elements from soil and division (Tahir and Karim, 2010).

Conclusion

The present findings have shown that irrigation with magnetized water could be employed as one of the most valuable modern technologies that can assist in saving irrigation water and improving yield and quality of Canola under newly reclaimed sandy soil. The usage of magnetic water in the agricultural production will enable intense and more quantities and qualitative production.

Conflict of Interests

The authors have not declared any conflict of interests.

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