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Review

Advances in carotenoid increments in storage parts of African staple crops

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The importance of vitamin A and other carotenoids in controlling micronutrient based deficiencies in particular VAD has been emphasized in recent years. This has resulted into demands for availing these nutrients in forms that are easily accessible for most of the populations in micronutrient deficient areas. Specifically in Africa, various programs have been instituted to bio-fortify crops with nutrients with more emphasis being put on Vitamin A fortification. Much as advances have been made in this area, a number of programs have registered little success while others have not taken off. In this review, advances in breeding for vitamin A increments are discussed. Countries where successes have been achieved are also highlighted while efforts in a number of areas for the different staple crops have been given due emphasis. In particular, breeding strategies have been discussed, and examples of successful breeding strategies highlighted to inform future efforts. In addition, the effect of processing on retention of vitamin A in processed products has been discussed with specific recommendations on identification of crop specific processing procedures. Such procedures should be optimized before adoption to allow for minimal losses in vitamin A and other related nutrients. We conclude that much as advances have been made, specific efforts are still needed in certain staples in order to provide benefits to the African consumers.

Key Words: African staples, Carotenoids, Malnutrition, Biofortification, Retention

INTRODUCTION

Vitamin A (retinol) is essential for vision, cell growth and tissue differentiation, and is critical for development during pregnancy and breastfeeding. Preformed vitamin A is found almost exclusively in animal products. Vitamin A content ranges from about 30 µg retinol per 100 ml in full cream milk up to as much as 16,000 µg of retinol per 100 g in the liver (Wijesundera et al., 2012). However, dependence on animal products is not sustainable in sub-Saharan Africa as such products are not affordable and are associated with major chronic syndromes such as cardio-vascular disease. As a result, there is need to provide precursors for vitamin A in adequate amounts in most staple crops. Carotenoids, especially α and β-carotene, which are potential vitamin A precursors are present in plants and plant products and some may contain up to 310 µg of β-carotene per gram especially in

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fruits and vegetables (Khoo et al., 2011). Carotenoids are biologically less available than retinol but highly abundant in plant sources than animal foods. However, fruits and vegetables are seasonal crops with variable availability (FIT-Uganda, 2006) and hence daily supply cannot be guaranteed as the consumption patterns of fruits and vegetables are uncertain. For instance, it was reported in Cincinnati city of Ohio that only 18% of adults eat the recommended daily serving of fruit or vegetable (Interact for Health, 2014). In a related study in Minnesota, USA, it was reported that 91% of males and 86% of females consumed less than 3 servings of fruit or vegetable per day despite reported availability by 89% of the subjects (Arcan et al., 2007). Similarly, certain population groups (prisoners and refugees) take restricted diets that exclude certain types of micronutrient rich foods. Such consumption patterns show that there is need for alternative measures for meeting vitamin A requirements in various populations.

Carotenoids (vitamin A precursors) are C40 polyenes which are essential in plant development, photosynthesis, root mycorrhizal interactions and production of phytohormones (Esuma, 2016). Carotenoids comprise a large isoprenoid family and most are 40-carbon tetra-terpenoids derived from phytoene (DellaPenna and Pogson, 2006). The carotenoid backbone is either linear or contains one or more cyclic β-ionone or ε-ionone rings or, less frequently, the unusual cyclopentane ring of capsanthin and capsorubin (Arimboor et al., 2015). Carotenoids exist in non-oxygenated forms referred to as carotenes or their oxygenated derivatives the xanthophylls (DellaPenna and Pogson, 2006). The most commonly occurring carotenes are β-carotene, stored in chloroplasts and lycopene in chromoplasts of some flowers and fruits and other plant storage organs. The most abundant xanthophylls (lutein, violaxanthin, and neoxanthin) occur in photosynthetic plant tissues as vital components of the light-harvesting complexes (DellaPenna and Pogson, 2006).

Carotenoid biosynthesis and storage in plants

Carotenoid pigments are produced by the isoprenoid biosynthesis pathway and uses isopentenyl pyrophosphate (IPP), a 5-carbon compound, as the building block. There are two biosynthetic routes for IPP biosynthesis: a) the major one occurs in the cytoplasm and utilizes acetyl-CoA units in sequential reactions that lead to formation of IPP; b) the second route occurs in the chloroplasts, whereby, IPP is formed through a reaction initiated by the condensation of pyruvate and glyceraldehyde-3-phosphate (G-3-P). This reaction is catalyzed by enzyme 1-deoxy-D-xylulose-5-phosphate synthase and leads to the formation of 1-deoxy-d-xylulose 5-phosphate (DXP). The DXP reductively isomerizes to form a 2C-methyl-d-erythritol-2, 4-cyclophosphate (MEP) in a reaction catalyzed by enzyme DXP reducto-isomerase. Finally, MEP is subsequently converted through a series of reactions to isopentyl-pyrophosphate (IPP) and dimethyl allyl pyrophosphate (DMAPP).

Thereafter, various isoprenoids can be formed through condensation of various units of IPP molecules. Typically, a molecule of IPP condenses with one molecule of DMAPP to form geranyl pyrophosphate (GPP) in a reaction catalyzed by GPP synthase. The next steps involve sequential addition of IPP molecules to form a 20-carbon compound, geranyl geranyl pyrophosphate (GGPP) (Figure 1). Two molecules of GGPP condense to form phytoene, in a reaction catalyzed by phytoene synthase (PSY) (Cunningham Jr and Gantt, 1998). Phytoene then undergoes a series of four desaturation reactions resulting into formation of lycopene (ψ, ψ -carotene). Lycopene is vital in synthesis of carotenoid compounds in plants.

Typically, lycopene is cyclized to form α-carotene and β-carotene, in a reaction catalyzed by enzyme lycopene β-cyclase (LCYB). The α-carotene can be further oxidized to form zeinoxanthin and leutin, while β-carotene can be oxidized to form either zeaxanthin, or violaxanthin, or neoxanthin, depending on the plant species (Cunningham Jr and Gantt, 1998; DellaPenna and Pogson, 2006). The carotenoids formed in synthetic cells especially in leaves can then be taken from the source organs to the sink via protein mediated translocative activities that are tightly regulated to reduce photo-oxidation. In staple crops, storage of such carotenoids is essential as a micronutrient to populations that depend on such staples. These provitamin A carotenoids from plants represent an additional major dietary source of vitamin A for most of the world's population (Weber and Grune, 2012).

Micronutrient malnutrition and Vitamin A deficiency (VAD)

Micronutrient malnutrition is a major underlying cause of health problems in developing countries. In particular, Vitamin A deficiency (VAD) can result either into night blindness or *Xerophthalmia* (dryness of the conjunctiva and cornea) and *Keratomalacia* (softening and ulceration of the cornea); causing total blindness (Semba, 1998; Gegios et al., 2010). The deficiency is more prevalent in children under 5 (Maziya-Dixon et al., 2006) and in pregnant mothers due to high nutrient demands by the fetus and mother. VAD accounts for over 600,000 deaths each year globally among children below 5 years of age (Hotz et al., 2012). Indeed, the World Health Organization (WHO) classified VAD as a public health problem in one third of children aged 6 months to 5 years in 2013. Of these, the highest rates were in sub-Saharan Africa at 48% and South Asia at 44% (Stevens et al., 2015; UNICEF,

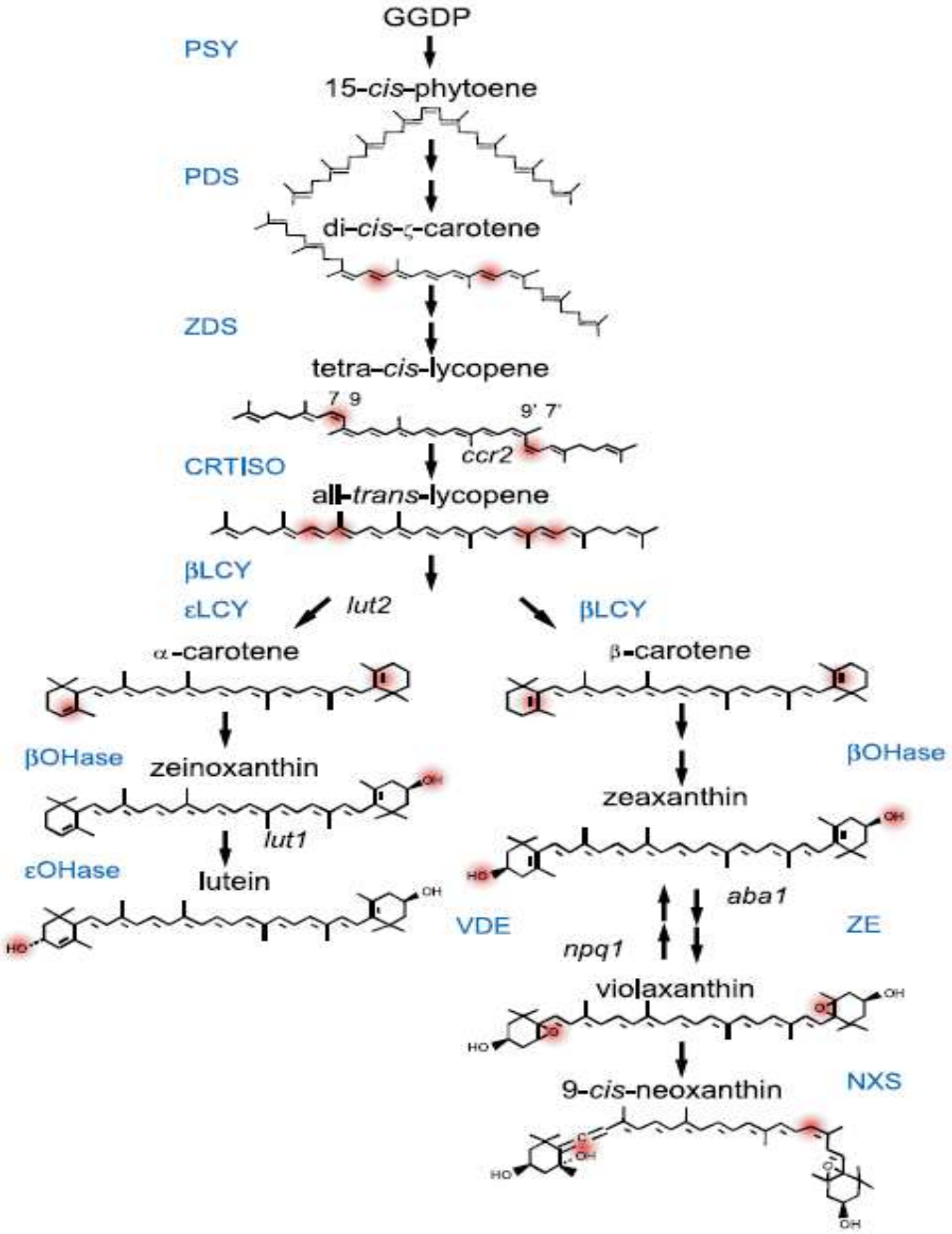

Figure 1. The biosynthetic process for carotenoids in plants: The pathway for carotenoids in plants. Adapted from (DellaPenna and Pogson, 2006). CRTISO, Carotenoid isomerase; β-LCY, β-carotene cyclase; βOHase, β-carotene hydroxylase; εLCY, ε-cyclase; εOHase, ε-carotene hydroxylase; NXS, neoxanthin synthase; PDS, phytoene desaturase; PSY, phytoene synthase; VDE, violaxanthin deepoxidase; ZDS, ζ-carotene desaturase; ZE, zeaxanthin epoxidase.

2016).

Beyond the role in vision, vitamin A plays a critical role in modulation of the immune function of the body. Notably, experimental observations in early 1920s and 30s led to the reputation of vitamin A as the “anti-infective” vitamin (Semba, 1998) which on supplementation reduced child mortality by 30% (Mora et al., 2008). Indeed, high-dose vitamin A supplementation had become recommended therapy for measles in developing countries and in the United States in the 1980s (Semba, 1998). Supplementation with vitamin A has also been shown to enhance resilience in AIDS

patients by increasing $CD4^+$ lymphocytes during HIV infection (Semba, 1998) and enhances immunity against cancer and HIV (Semba, 1998; Mora et al., 2008).

FORTIFICATION OF STAPLE FOODS FOR PROVITAMIN A ENHANCEMENT

Food fortification refers to addition of an essential nutrient to a food (Allen et al., 2006). The success of any food-fortification programme is the improvement in the nutritional and health status of a targeted population (Wirakartakusumah and Hariyadi, 1998). In line with this, the fortified food should be accepted and consumed by the targeted population. Thus, factors such as quality, taste, and cost of the fortified products play important roles in determining the effectiveness of the fortification programmes. There are two approaches of food fortification: conventional fortification, involving addition of nutrients during food processing, and biofortification, where a plant carrier is modified to express the added nutrient during growth.

Conventional food fortification

Conventional food fortification is the addition of essential nutrients to a food during food processing in appropriate concentrations that ensure accuracy and consistency (Organization, 2016). This depends on a well-functioning dosing technology and a reliable method of detection. The process involves the use of a food carrier (food to be fortified) to which the target nutrients are added either singly or in a premix (cocktail of target nutrients). Following the fortification process, detection of the added nutrient is carried out to confirm presence of the nutrient, in the desired quantities.

Conventional food fortification is particularly effective in tackling deficiencies, especially in densely populated urban areas where land for cultivation of food crops is scarce (Triggle, 2004). This method of nutrient enhancement is also attractive because it does not require the target groups to change their diet but can be implemented by the food industry and because it reaches large numbers of consumers through retail (Triggle, 2004). World Bank studies suggested that the annual per capita cost of fortifying a food with vitamin A is between USD 0.69 USD and USD 0.98 per capita per year (Triggle, 2004). Conventional fortification would therefore be a more cost-effective method for impacting on vitamin A intake of communities, especially in urban areas.

However, the implementation and effectiveness of this method to alleviate vitamin A deficiency in populations that depend on own-saved food is difficult. Such programs are affected by other socio-economic factors that influence dependence on major staple foods. In particular, the subsistence form of agriculture, coupled with the pressing socio-economic demands such as education and health care compel the resource-poor farmers to sell off fruits, animals and animal products such as eggs in order to earn a living. Moreover, the cost of fortified foods is high and usually unaffordable for such farmers due to high poverty levels (Esuma, 2016; Iannotti et al., 2013). Therefore, there is always need to search for alternative dietary intervention methods so as to have a wider coverage, especially in vulnerable groups in rural areas.

Biofortification of staple food crops with carotenoids

Biofortification refers to the breeding and genetic modification of plants so as to improve their carotenoid content. It differs from conventional fortification in that biofortification increases nutrient levels in consumable crop storage organs during plant growth, rather than during processing of the crops (Organization, 2016). In this regard, the biofortified crop should be able to express the carotenoids during its growth period. Biofortification can be either by genetic engineering, normally referred to as modern biotechnology, or by conventional plant breeding.

Genetic engineering approaches have been used to either increase or modify the carotenoid content in plants through manipulations in the carotenoids biosynthetic pathway. Phytoene synthase (PSY) is a branching enzyme that directs substrates irreversibly to carotenoids. Hence, it has been the target in several genetic manipulation studies (Naik et al., 2003). For instance, the constitutive over-expression of PSY in plants that do not normally produce carotenoid pigments; as in tobacco, tomato and rice, led to substantial increase in carotenoid accumulation (Naik et al., 2003; Fraser et al., 2002; Paine et al., 2005). Similarly, the genetic manipulation of rapeseeds (*Brassica napus*) using a bacterial PSY gene (*crtB*) to increase carotenoid content resulted in up to a 50-fold increase in carotenoids, α- and β-carotenes being the predominant ones. Similarly, the PSY gene was also cloned into rice (Burkhardt et al., 1997; Ye et al., 2000) to induce carotenoids synthesis and storage, the case of the Golden Rice. In addition, DNA constructs aimed at upregulating the expression of lycopene β-cyclase gene were introduced in tomatoes and transformants showed significant increments in carotenoids content (Naik et al., 2003).

Conventional plant breeding refers to the crossing of plants with relevant characteristics, to form a crossbreed that exhibits inherited traits from both parent plants (Royal Society, 2016). This is followed by selection and multiplication of the offspring with the desired combination of characteristics. Depending on the crop, the conventional breeding process may take 10 or more years before a variety can be released to the grower (Caligari and Forster, 2012) and involves step-wise

procedures leading to variety release (Fukuda and Saad, 2001; Esuma, 2016). Usually, farmers are involved in the final stages, especially at farm level testing (Esuma, 2016). Modified and shorter breeding procedures have been made for various crops (Kawuki et al., 2011; Pfeiffer and McClafferty, 2007) by modifying and/or eliminating specific steps in the conventional breeding process or by adopting molecular selection methods such as marker-assisted selection, marker-assisted recurrent selection, or marker-assisted back-crossing (Moose and Mumm, 2008). Other efforts such as the double haploid technology (DH) with potential improvement to the conventional schemes are also being developed. Currently, breeders are optimizing genomic selection tools using whole-genome coverage markers such as single nucleotide polymorphism (SNP) to develop prediction models which enormously reduce the breeding cycle and the number of hybrids to be evaluated in the field (de Oliveira et al., 2012).

Biofortification is advantageous in addressing micro-nutrient deficiency due to its long-term cost-effectiveness and its ability to reach underserved, rural populations. The upfront investments in plant breeding yields micronutrient-rich biofortified planting material for farmers to grow at virtually zero marginal cost. These can be evaluated and adapted to new environments, multiplying the benefits of the initial investment with minimal recurrent expenditures (Bouis and Saltzman, 2017). Biofortified crops can easily reach rural populations who have limited access to diverse diets or other micronutrient interventions. Target micronutrient levels for biofortified crops are set to meet the specific dietary needs of women and children, based on existing consumption patterns (Bouis and Saltzman, 2017; Mwanga et al., 2016). Subsequently, biofortification may present a way to reach populations with preferred food traits especially where supplementation/conventional fortification activities may be limited (Organization, 2016). Accordingly, global efforts have been tailored to biofortification of staple food crops to help alleviate deficiencies associated with overdependence on the foods. Major foods under biofortification programmes in Africa include sweet potato, cassava, maize, and banana.

CURRENT STRATEGIES FOR INCREASING CAROTENOID CONTENTS IN STORAGE PARTS OF THE PLANT

The elucidation of carotenoid biosynthesis pathway and genes involved in the control and regulation of the pathway is important in breeding for increased carotenoid content. This process can be hampered by: 1) the fact that synthesis of β-carotene is induced by GGPP, a metabolic precursor for other vitamins and pigments whose synthesis could be decreased; 2) interference with the well-balanced regulatory mechanism of the pathway and 3) the need for highly lipophilic nature of carotenoids provision of storage in plants. Therefore, high carotenoid production should focus on increased precursor supply, maintaining the balance between interacting metabolic pathways and targeting of tissues that are capable of incorporating lipophilic molecules (Naik et al., 2003). Also, increased levels of carotenoids in storage parts of higher plants might be due to down-regulation metabolite synthesis. Even then, efforts have been put forward to breed and increase provitamin A content in storage parts of major staple food crops in Africa (Table 1 and Figure 2).

Plant breeding has mainly been used to increase the carotenoid content in edible tissues of crops including non-photosynthetic storage parts. It has been observed that such processes do not cause any interruption to the plant, and that the pro-vitamin A levels can be influenced to attain the required thresholds. Once such plant varieties have been attained, rigorous promotion and awareness creation is then required for adoption. Such success has been attained with the Orange Sweet Potato (OSP) in tropical and subtropical Africa and can be easily applied to other African staples such as cassava, maize and bananas that are nutritionally superior, well adapted to local growing conditions and more profitable for farmers. However, the bioavailability of plant provitamin A varies widely in relation to the food crop, genotype (including sink activity of the storage organ) cooking method, individual genetic factors and consumption of fat with the meal. Other factors such as post-harvest food storage, processing environment and general food handling also affect carotenoid availability. Thus, optimisation of such factors for increased bio-availability is of paramount importance.

Breeding strategies for enrichment of Vitamin A in storage parts staple crops

Vitamin A enhancement in storage parts of staple crops has been achieved through a range of breeding approaches including transgenic, agronomic and conventional procedures. In the case of carotenoids, enrichment within the plant has been achieved through expression or up regulation of genes and or gene products involved in carotenoid synthesis (DellaPenna and Pogson, 2006). In a number of instances as related to transgenesis, genes can be added, removed or altered in such a way that the production and hence translocation of carotenoids from source to sink organs is enhanced. However, in all cases, care should be taken that the product from such manipulations is accepted by the consumers.

Biofortification as a strategy combines the use of both conventional and sometimes transgenic approaches for accumulation of carotenoids in storage parts (Bouis and Welch, 2010). In biofortification, the breeders aim at

Table 1. Major staple food crops in Africa, that have been biofortified for increased provitamin A content and their distribution.

Crop	Sweet potato (OFSPs)	Cassava (Yellow cassava)	Maize (orange maize)	Banana (Golden banana)
Varieties released	NASPOT 12 O, NASPOT 13 O, Kakamega (SPK004) Ejumula, NASPOT 9 O, NASPOT 10 O, NASPOT 7, NASPOT 8 Dimbuka- Bukulula, Resisto, Persistente, Tiba, LO-323	I011661 in DRC UMUCASS 36, UMUCASS 37, UMUCASS 38, TMS07/0593, TMS07/0539 TMS07/0220	MeruVAH517 MeruVAH519	Local varieties in DRC and Burundi, having appreciable levels are available
Level of pVAC (wild types)	30-100 ppm	0-19 ppm	0-19 ppm	1.4-11.3 ppm
Target level (pVAC)	32 ppm	15 ppm	15 ppm	Up to 20 ppm
Other variety characteristics	Yield = 60t/ha on station and 12t/ha farmer field, DMC (30-35%), moderate resistance to disease and pests	Yield, early maturity, tolerance to pests and diseases, DMC, pound-ability, mealiness, sweetness, ease of peeling, marketability, and in-ground storage	higher zinc content, competitive grain yield and consumer preferred end-use quality traits	No specific carotene rich varieties released so far
Country of release	Uganda, Mozambique. High adoption in all countries in SSA	Nigeria, DRC, Kenya, Ghana, Sierra Leone, Malawi	DRC, Ghana, Malawi, Mali, Nigeria, Rwanda, Tanzania, Zambia, and Zimbabwe	Trials in Australia and in East Africa
Target countries	East and Southern Africa	West Africa, East Africa and part of south Africa	Sub Saharan Africa and South Africa	East Africa, DRC
References	Mwanga et al. (2007), Mwanga et al. (2016), Laurie (2001)	Esuma (2016), Ssemakula and Dixon (2007), Ssemakula et al. (2008), Njoku et al. (2014)	Menkir and Maziya-Dixon (2004), Pixley et al. (2013), Ortiz et al. (2016)	Ekesa and Nabuuma (2016), Paul et al. (2017), Fungo and Pillay (2011), Mbabazi (2015)

providing mechanisms for accumulation of carotenoids in addition to reduction of ant-nutrient substances that inhibit carotenoid bioavailability after consumption. Relatedly, breeders can increase particular substances in the crop that stimulate and promote carotenoid bioavailability in storage parts or the crop product. If this is not done, then the breeding process would not yield products that are of use to consumers. This calls for a thorough appraisal of the agronomic performance of the crop which should be enhanced to allow for sustained crop performance (Welch, 2002).

As a strategy, transgenic approaches are important in crops where the carotenoids are absent or do not occur in such significant amounts as would be increased through conventional means (Bouis and Saltzman, 2017). This approach is precise in delivering significant amounts of nutrients to the crop in question and tends to shorten the breeding cycle. In addition to increasing carotenoid contents, the approach also ensures that specific agronomic and performance properties of the crop (especially sink size and activity) are maintained. However, the approach is affected by the highly risk averse regulatory approval processes as has been seen in the case of “Golden rice” (Wesseler and Zilberman, 2014).

Successful biofortification strategists like Harvest Plus have used specific approaches based on conventional breeding approaches. Such approaches are indeed faster and better means of getting the carotenoid rich crops to consumers.

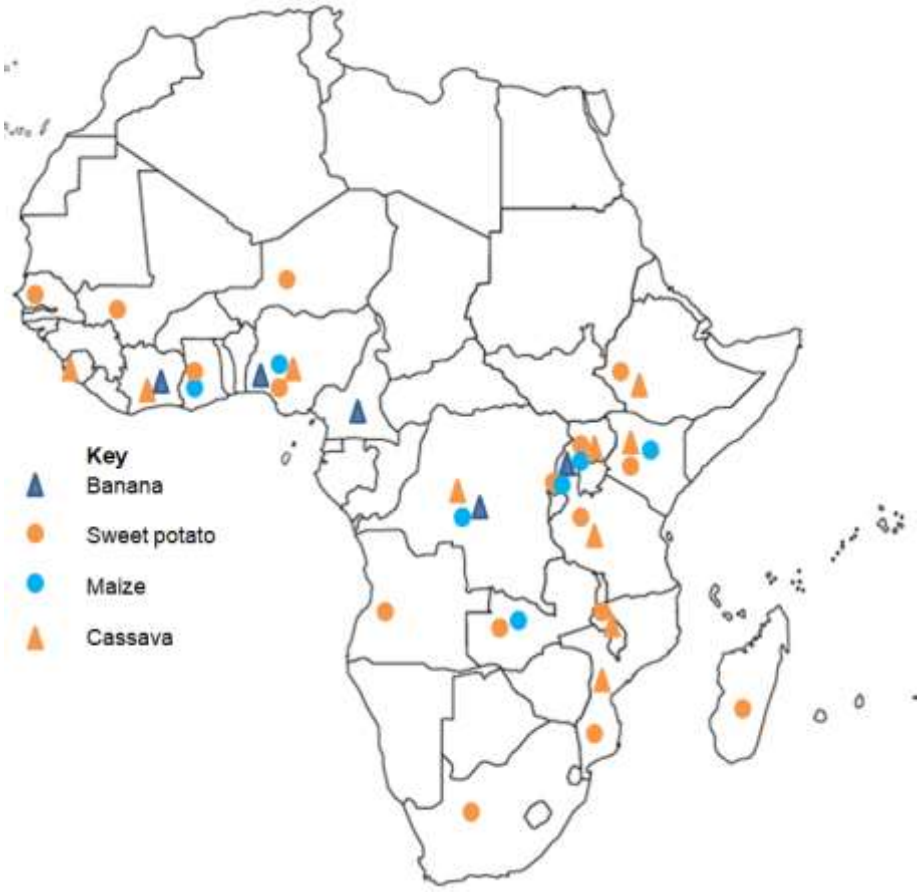

Figure 2. Map showing major staple food crops under biofortification for provitamin A enhancement in Africa and the countries involved in biofortification by 2017.
Source: Adapted and modified from HarvestPlus (2014).

The approaches involve the identification of varieties that are already adapted to a particular location of interest but also carry "significant carotenoid content". Such varieties are then packaged for release and/or dissemination as "fast track" varieties. This provides spot on solutions to populations in need and can complement the long and laborious carotenoid conventional breeding processes. In addition, harvest plus conducts a range of multi-locational trials across specific similar geographical locations that allow for testing and faster release of developed nutrient rich varieties (Bouis and Saltzman, 2017).

Therefore, it is important to note that conventional breeding can be coupled to modern genetic approaches for enhanced carotenoid increments in storage parts of staple crops. However, the application of genetic engineering approaches in delivering such products is still elusive. Thus the breeding of African staples for enhanced carotenoid concentrations has been based on conventional approaches. Such approaches have delivered important crops such as sweet potato, cassava, and maize that contain higher levels of provitamin A carotenoids (pVACs) (Table 1). They have also been used to deliver other micronutrient rich crops such as beans and a number of cereals.

Vitamin A biofortified crops for consumers in Africa

Sweet potato

Sweet potato is widely consumed in sub-Saharan Africa and was the first biofortified crop developed and released by the International Potato Center (CIP), HarvestPlus and their partners. It accumulates provitamin A up to 100 ppm exceeding the target level of 32 ppm (Andersson et al., 2017). The primary evidence for the effectiveness of biofortification in accumulation of carotenoids in storage parts comes from Orange Sweet Potato (OSP). In Uganda, orange-fleshed landrace cultivars named 'Ejumula' and 'SPK004' (Kakamega) (Mwanga et al., 2007), and developed varieties (NASPOT 9 O' (NASPOT 10 O, NASPOT 12 O and NASPOT 13 O) with yellow roots (sign of pVACs accumulation), were released in 2004, 2007 and 2013 (Mwanga et al., 2016). Biofortified

OSP varieties have been released in more than 15 countries across sub-Saharan Africa with a record adoption rates greater than 60% above control communities (Bouis and Saltzman, 2017). Introduction of these varieties resulted in increased vitamin A intakes among children and women, improved vitamin A status among children and decreased the prevalence of low serum retinol by 9% points (Mwanga et al., 2016). Women who consumed OSP also had a lower likelihood of having marginal vitamin A deficiency. Recent research on the health benefits of biofortified OSP in Mozambique showed that biofortification improved child health as indicated by reduced prevalence and duration of diarrhoea in children under five years of age (Bouis and Saltzman, 2017).

Cassava

Cassava is a dietary staple in much of tropical Africa, and grows well in poor soils with limited labour requirements. Total carotenoid concentration in fresh yellow cassava is primarily in the form of all-trans-β-carotene and is located in the parenchyma cells, the storage cells of the roots (Talsma, 2014). Breeding programmes for provitamin A cassava such as the International Center for Tropical Agriculture (CIAT) and the International Institute of Tropical Agriculture (IITA) generate high-provitamin A sources via rapid cycling in pre-breeding and provides in-vitro clones and seed populations for local adaptive breeding. Indeed, the breeding efforts at CIAT have already led to the generation of cassava genetic stocks that have accumulated up to 25 µg/g of β-carotene in fresh roots (Ceballos et al., 2013). In Nigeria, three first-wave provitamin A cassava varieties with 6–8 ppm of provitamin A (about 50% of the target) were released in 2011 followed by three other varieties with up to 10 ppm (66% of the target) in storage roots released in 2014. However the breeding target is to deliver varieties with up to 15 µg/g fresh weight of carotenoids in fresh roots (Talsma, 2014). National programmes have also released yellow cassava varieties in Ghana, Malawi, and Sierra Leone, and regional trials are underway for fast-tracking release in other countries in West and East Africa that have similar agro-ecologies. In Uganda, elite provitamin A cassava germplasm was introduced from CIAT and IITA around 2012 (Esuma et al., 2012) for evaluation under local field conditions. Breeding efforts have currently given rise to varieties with carotenoids content up to 12 µg/g, awaiting release for farmer adoption (Esuma et al., 2012). As earlier stated, this is affected by the negative correlation between carotenoid increments and dry matter accumulation hence the need to combine high carotenoid content and high dry matter content of biofortified germplasm for Africa. As earlier stated, adoption of provitamin A varieties by farmers is hindered by the negative correlation between carotenoid content and dry matter content, hence the need to improve the dry matter content of biofortified germplasm for Africa.

Maize

Maize is the most important cereal crop in sub-Saharan Africa and is also an important staple in Latin America. Initial screening of more than 1,500 maize germplasm accessions found ranges of 0–19 ppm provitamin A in existing maize varieties, exceeding the provitamin A target of 15 ppm (Menkir et al., 2014; Andersson et al., 2017). Provitamin A maize breeding programs at the International Maize and Wheat Improvement Center (CIMMYT), IITA, and the Zambia Agriculture Research Institute (ZARI) began in 2007. Both hybrid and open-pollinated (synthetic) biofortified varieties are being developed for with improved carotenoid storage in the grain (Andersson et al., 2017). In Africa, more than 40 provitamin A maize synthetic hybrids, single-cross hybrids, and three-way hybrids have been released in the DRC, Ghana, Malawi, Mali, Nigeria, Rwanda, Tanzania, Zambia, and Zimbabwe (Andersson et al., 2017). The first wave of varieties released in 2012/2013 contained 6-8 ppm additional provitamin A (about 50% of the target increment) in the dry grain, while second-wave varieties (released in 2015/2016) contained about 10 ppm additional provitamin A (66% of the target increment). Varieties that fully meet the provitamin A target level are being tested in multi-location trials across sub-Saharan Africa and are expected to be released in 2018 (Andersson et al., 2017). All biofortified varieties combine competitive grain yield and consumer preferred end-use quality traits with higher provitamin A content.

Banana

Banana is an important staple food and source of income for over 100 million people in Sub-Saharan Africa, with consumption averaging 300 kg per person per year in the East African highlands and the Great Lakes region of Africa (UNSCT, 2007). The high consumption rate makes banana an important source of carbohydrates, vitamins and minerals in the diets of these populations (Davey et al., 2007). However, most of local cultivars have significantly lower levels of pro-vitamin A in the fruit and are consumed in a region where VAD deficiencies range from 39-50% (IFPRI, 2016) and way beyond the WHO acceptable intervention level of 15% (WHO, 2009). Thus the inherent potential of these cultivars for improvement into pVAC-rich cultivars with organoleptic properties that compare well with that of local cultivars must be harnessed.

Evaluation of some of the banana genotypes have already shown a wide variation in provitamin A

carotenoids (pVACs) content with values as high as 220 nmol g^{-1} dry weight (DW) (Davey et al., 2009). Other studies by Ekesa et al. (2013) in popular banana cultivars in Eastern Africa reported pVAC ranges from 7 to 27 nmol g^{-1} DW. These can be improved using germplasm from other sources that have higher levels of pVACs than local cultivars (Ekesa et al., 2013; Englberger et al., 2003; Fungo and Pillay, 2011). On the basis of consumer reliance on banana for food and the high bio-accessibility of vitamin A in banana (Ekesa et al., 2013), pVAC-rich banana cultivars form a vital route that answers to the high levels of prevalence of VAD within Eastern Africa.

Notwithstanding efforts for biofortification of banana with provitamin A carotenoids in Africa, the most outstanding successes are still on the proof of concept. PVA-biofortified transgenic Cavendish bananas were generated in collaboration with African partners and field trialed in Australia with the aim of achieving a target level of 20 lg/g of dry weight (DW) b-carotene equivalent (b-CE) in the fruit (Paul et al., 2017). However, these have not been deployed in the greater banana region of east Africa. Needless to say, it is evident that a shift from low-carotenoid to high-carotenoid banana cultivars would lead to increased vitamin A content of the diet and thus possibly lead to improved vitamin A status among consumers.

RETENTION OF CAROTENOIDS AND VITAMIN A DURING FOOD PROCESSING

Staple crop storage parts processing results into reduction in carotenoids content. The reduction in carotenoids content during processing differs: 1) from variety to variety (Chavez et al., 2007; Vimala et al., 2011); 2) from one processing method to the other (Vimala et al., 2011; Chavez et al., 2007) and 3) from different positions of the same storage part within the variety (Talsma, 2014). Talsma (2014) attributed the variations in carotenoids reduction among different positions within the same variety to the variable distribution of dry weight matter within a particular storage part. Carotenoids retention in staple crops products may vary from as low as 10% for heavily processed and roasted food granules, to about 87-90% in less processed storage parts (Ceballos et al., 2012; De Moura et al., 2015).

Generally, increased temperature and light conditions severely reduce the amount of available carotenoids. Reduced retention is mainly through a number of degradative pathways including the reaction of carotenoids with atmospheric oxygen (autooxidation), light (photodegradation) and heat (thermal degradation). Degradation can also be as a result of interactions of carotenoids with singlet oxygen, acid, metals, and free radicals within the product processing environment.

Given the complex nature of food based material matrix from biofortified crops, the retention/degradation of carotenoids is a complex process that is as a result of a range of factors. Carotenoid interaction with other materials/biomolecules within the food is rather not well understood (De Moura et al., 2015). However, various studies have focused on the understanding of carotenoid retention on the effect of external factors/processes on carotenoid availability. Thus it remains to be understood, on the exact physiological processes that take part in the degradation of carotenoids and hence reducing carotenoid retention.

From the review of different studies undertaken on a range of biofortified crops deployed especially in sub Saharan Africa (De Moura et al., 2015), retention levels have been elucidated and henceforth, recommendations on the same can be made (Figure 3). It was shown that in sweet potato, carotenoid retention is high if the food matrix is processed by boiling and/or production of porridge. Such processes retain carotenoid content well above 90% and would be ideal for the African settings. In maize, the best method of processing was making of porridge (80% retention) while in cassava, boiling and *fufu* production were scored as best method for high retention (over 90%). Among such boiling procedures as the most common form of food preparation, boiling with minimal water (half full) resulted into higher retention for carotenoids. Most of the biofortified crops in Africa are primarily processed by drying. Among the drying procedures, it was realised that shade and solar drying resulted in lesser degradation and hence higher carotenoid retention. Since most farmers store their produce after harvest, the recommended storage procedure among different crops that retains higher levels of carotenoids was storage in jute bags. It was also realised that the storage of the food material in the dark gave better results. Such information is critical in deployment of biofortified crops if at all, their intended benefits are to be realised by the intended beneficiaries.

CONCLUSIONS

Like other plants, African staple crops accumulate carotenoids through specified biosynthetic processes. However, for most crops that have consumer-accepted attributes, the level of carotenoid accumulation is low and may not cater for the nutritional requirements of the consumers. Through biofortification, staple crop carotenoid contents can be significantly improved as has been demonstrated in sweet potato, cassava, bananas and maize. Such improvements take into account the apparent variability in crop carotenoid contents and hence varietal improvements would best target varieties or cultivars that already contain appreciable amounts of carotenoids. On the other hand, such varieties may not carry specific consumer preferred traits. Hence, African breeding programs have created breeding pipelines for

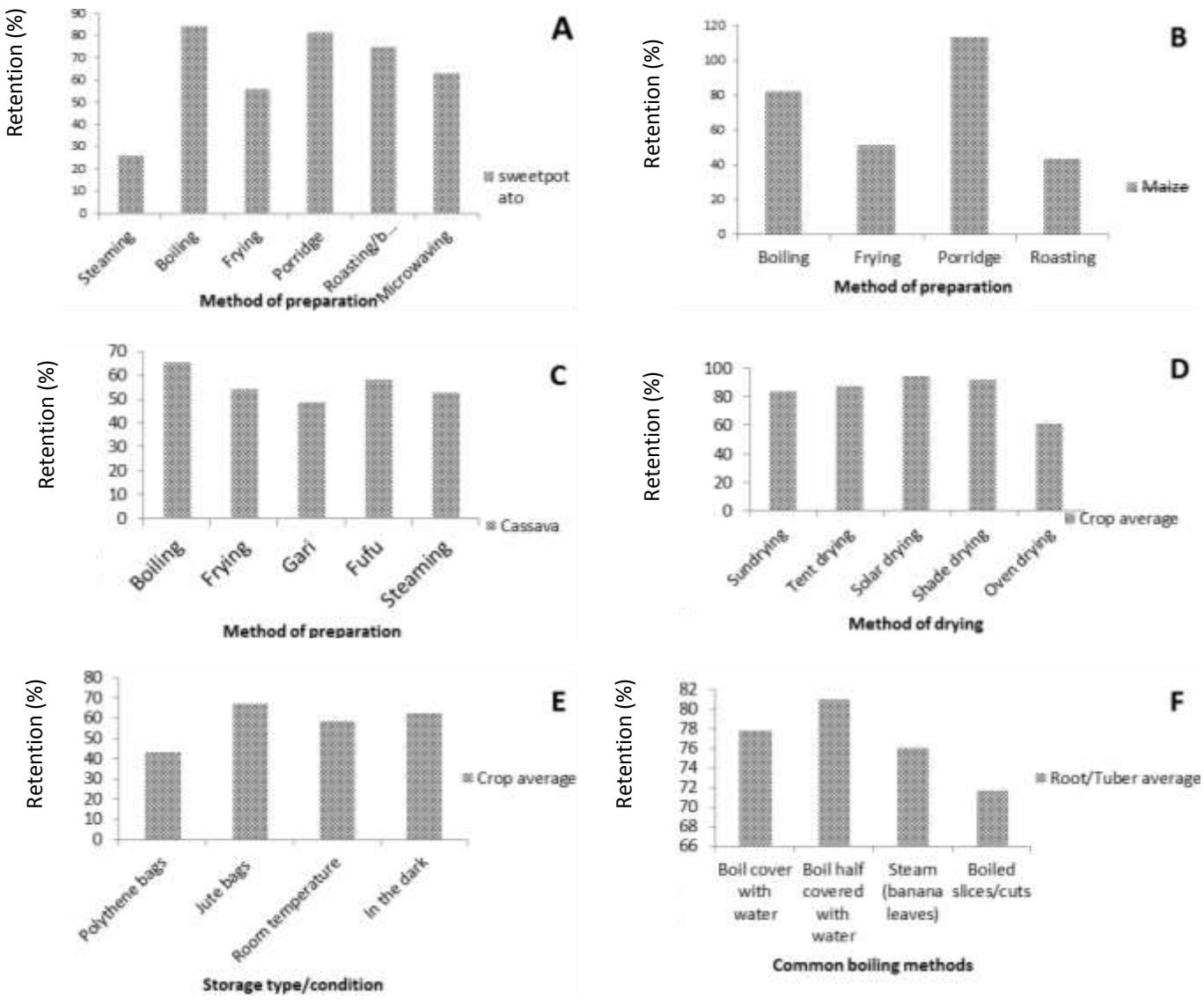

Figure 3. Retention of carotenoids in crops under different conditions. A= Average Carotenoid retention in Sweet potato; B=Average Carotenoid retention in Maize; C=Average carotenoid retention in cassava D=Crop average for carotenoid retention in various drying methods; E=Crop average for carotenoid retention during storage in bags or different light conditions; F=Root/Tuber average carotenoid retention during boiling.
Source: Adapted from De Moura et al. (2015).

the production of vitamin A rich crops.

These breeding pipelines have produced a range of varieties which have been adopted by farmers in different parts of Africa. The efforts have been very successful especially in sub Saharan Africa with almost all the countries having biofortified crop varieties. However, challenges still remain such as accumulation of the right amounts of carotenoids in storage parts to allow bio-accessibility, acceptability of the crop or their products, the susceptibility of such crops to diseases and pests and the retention of such carotenoids within the processed products meant for consumption.

Such challenges indicate the need for concerted efforts in breeding, food science, post-harvest technology and other in providing for highly bio-accessible, highly retainable and useful forms of these nutrients in staple crops. This would take the form of the clear understanding of the molecular and physiological aspects of carotenoid accumulation in crops plants. It would also require specific solutions related to appropriate processing procedures for these crops coupled with an understanding of the interaction of carotenoids with other molecules in the food matrix. Solutions to a range of challenges related to utilisation of biofortified would henceforth result into their

increased utilization and possibly reduce the rampant micronutrient related disorders in Africa.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Full Length Research Paper

Genetic Diversity Analysis of Ethiopian Elite Chickpea (*Cicer arietinum L.*) Varieties Based on Agronomic Characters

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This research aim to assess morphological diversity of the elite chickpea (*Cicer arietinum* L.) varieties in Ethiopia. Nineteen elite varieties of chickpea in Ethiopia were used to analyze the means and components of variability (genetic, phenotypic and environmental), and interrelationships (genetic and phenotypic) for yield and various other yield components. Such nineteen varieties were planted by the technique of Randomized Complete Block Design (RCBD) and three replications were used. Each genotype was sown in four rows with 4.8 m^2 (1 m x 4.8 m) plots area, with 40 cm and 1 m spacing between plots and blocks, respectively. In each plot, one hundred and sixty seeds were planted, using 10 cm spacing between plants These nineteen elite varieties of chickpea were evaluated for the traits of hundred seed weight, biological yield, grain yield, plant height, days to 50% flowering, number of primary branches, number of secondary branches, number of pods per plant, number of seeds per plant, harvest index and days to 90% maturity. Genetic variations were evident among released chickpea cultivars as confirmed by high phenotypic and genotypic variations for quantitative and qualitative traits. Analysis of variance revealed significant differences among the genotypes for all the characters except hundred seeds weight, days of 50% flowering and grain yield. Strong and positive significant correlation was observed between grain yield, biological yield, number of seeds per plant, number of pods per plant and number of primary branches; showing that their improvement led to yield improvement in chickpea. The result suggested from the mean values of number of seeds per plant, number of pods per plant and days of maturity that chickpea genotypes ICCV-14808, Mariye and ICCV-92069 may be used as parents in further breeding program to develop high yielding cultivars. Principal component analysis revealed that quantitative traits contributed a lot to chickpea genetic variability.

Key words: Agronomic characters, correlation coefficients, elite, principal component analysis, variation

INTRODUCTION

Chickpea (*Cicer arietinum* L.) is the third most important pulse crop in the world and it is an important cool season

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grain, self-pollinating legume crop, and it is a basic component of the human diet in many countries (FAOSTAT, 2009). The leading chickpea growing countries in the world are India, Pakistan, Mexico, Turkey, Ethiopia and Myanmar (Keneni et al., 2011). The crop most probably originated from the area of present day Southeastern Turkey and the adjoining areas of Syria (Harlan, 1992). India and Ethiopia have been proposed as secondary centers for diversity of cultivated chickpea (Harlan, 1992). However, Zeven and de Wet (1982) suggested that chickpea has different secondary centers of diversity located in at least four regions: the Near East Region (Comprising the Fertile Crescent); Hindustani Region (basically the current India and East Pakistan); Central Asian Region (with Afghanistan, Western Pakistan, Iran and the southern part of the former USSR); and the Mediterranean Region (including Lebanon and Palestine) (Talebi et al., 2008). Plant genetic resources and the genetic diversity present in them provide an assurance for future genetic progress and an insurance against unforeseen threats to agricultural production (Hari et al., 2008). The studies of genetic diversity of plants are very important for developing high yielding varieties and for maintaining the productivity of such varieties in the plant breeding strategies. The screening and selection for crop improvement would be based more likely on availability of promising genotypes; which solemnly depends on the availability for better agronomic traits coupled with disease resistance, earliness and high yield (Keneni et al., 2011).

Chickpea is the cheapest and readily available source of protein, fats and carbohydrates (Choudhary et al., 2012). Unfortunately, despite its nutritional values and economic importance, chickpea production is very low per hectare in the country (Ethiopia) (Bejiga et al., 1996). This is primarily due to poor genetic makeup of the available cultivars. Genetic variability is a prerequisite for any breeding program, which provides opportunity to a plant breeder for selection of high yielding genotypes. One way to estimate the genetic diversity is based on morphological traits which are the classical methods to distinguish variations based on the observation of the external morphological differences in different geographical regions (Ghaffari et al., 2014, Vienne et al., 2003; Hari et al., 2008). It is the earliest genetic marker used for assessment of variation and still has great importance. Moreover, morphological characters are simple to score and economical to use. In the studies of Ethiopian chickpea morphological characters, the landraces showed considerable variability within and between chickpea populations (Bejiga et al., 1996; Feven, 2002; Melese, 2005). However information on the associations between yield and its various components provide the basis for the selection of improved varieties. The objective of this study is to assess morphological diversity of the elite chickpea varieties of Ethiopia, using quantitative characters of the chickpea varieties.

MATERIALS AND METHODS

Studying site

The experiment was conducted in Ethiopia at Debre Zeit Agricultural Research Center (DZARC), which is located in East Shoa zone of Oromia regional state; 47 km in the direction of South East of capital Addis Ababa. The geographic location of DZARC is 8°44’N latitude and 38°58’E longitude, with an elevation of 1860 m.a.s.l. The research center receives an annual rainfall that ranges from 452.8 to 934.2 (ml), with annual mean of 691.5 ml. The temperature of this location ranges from 10.76°C to 27.83°C, with mean annual temperature of 19.32°C. The dominant soil types of DZARC are Vertisols, Mollisols and Alfisols (DZARC, 2009; Melese, 2005).

Experimental materials

Nineteen elite chickpea genotypes were grown at DZARC. All the agronomic practices were carried out throughout crop growing season. The description of the nineteen genotypes along with their origin/source is given in (Table 1).

Experimental design and layout

A Randomized Complete Block Design (RCBD) with three replications was used. Each genotype was sown in four rows of 4.8 m^2 (1 m x 4.8 m) plots area and 0.4 m and 1 m spacing between plots and blocks, respectively. In each plot, one hundred and sixty seeds were planted by using 0.1m spacing between plants. Ten individual plants were tagged randomly from each genotype per plot and used for morphological data recording and the following qualitative and quantitative agronomic characters or morphological traits were recorded, using IBPGR descriptors (IBPGR, 1993). Data were recorded for each variety on a number of days to 50% flowering (DTF) and was recorded at the time when at least 50% plants showed the appearance of first flower. Days taken to maturity (DTM) were calculated from the date of planting to the date when 90% plot turned brown and ready for harvest. Maturity data were recorded for hundred seed weight (HSW), grain yield (GY), plant height (PHT), Number of pods per plant (NPP), Number of seeds per plant (NSP), number of primary branches per plant (NPB), number of secondary branches per plant, biological yield (BYD), and harvest index (HI). For data on plant bases, the mean of ten plants which were randomly selected from the two central rows for the plot bases and the two interior rows were used for data collection.

Statistical procedures

Analysis of Variance (ANOVA) was studied using according to Gomez and Gomez (1984) using SAS (1999) for calculating genotypic, phenotypic and environmental variation components and Least Significant Difference (LSD) test was used for pair wise comparison of means. ANOVA was computed for all quantitative traits to detect the variability present among the nineteen elite chickpea varieties. The analysis of variances was carried out following the standard procedure which is applicable to randomized block design as suggested by Gomez and Gomez (1984), using SAS (1999) statistical computer software. The variation of each morphological trait such as quantitative traits was estimated using simple statistical measures: mean, range, genotypic and phenotypic variances and coefficient of variations. The phenotypic and genotypic variation and coefficient of variations were calculated following the formula suggested by Singh and Ocampo (1977).

Table 1. Details of chickpea (*Cicer arietinum* L.) varieties used for diversity analysis.

S/N	Variety Code	Year of release	Variety name	Type
1	DZ-10-11	1974	DZ-10-11	*Desi*
2	Dubie	1978	Dubie	*Desi*
3	Mariye	1985	Mariye	*Desi*
4	ICCL 82104	1994	Worku	*Desi*
5	ICCL 82106	1995	Akaki	*Desi*
6	ICCV-92033	2005	Kutaye	*Desi*
7	ICCV-92006	2006	Mastewal	*Desi*
8	ICCV-92069	2006	Fetenech	*Desi*
9	ICCX-910112-6	2007	Natoli	*Desi*
10	Minjar	2010	Minjar	*Kabuli*
11	DZ-10-4	1974	DZ-10-4	*Kabuli*
12	FLIP 89-84C	1999/2000	Arerti	*Kabuli*
13	ICCV-93512	1999/2000	Shasho	*Kabuli*
14	ICCV-92318	2004	Chefe	*Kabuli*
15	FLIP 88-42C	2004	Habru	*Kabuli*
16	FLIP-97-63C	2005	Ejere	*Kabuli*
17	FLIP-97-66C	2005	Teji	*Kabuli*
18	ICCV-14808	2006	Yelibe	*Kabuli*
19	Monino	2009	Monino/Acos Dubie	*Kabuli*

From the analysis, phenotypic variance, genotypic variance, phenotypic coefficient of variation (PCV), and genotypic coefficient of variation (GCV) were calculated:

$$PCV = (\sqrt{\sigma^2 p} / \bar{x}) \times 100$$

Where, PCV= phenotypic coefficient of variation and $\bar{x}$ = Population mean.

$$GCV = (\sqrt{\sigma^2 g} / \bar{x}) \times 100$$

Where, GCV= genotypic coefficient of variation and $\bar{x}$ = Population mean.

RESULTS AND DISCUSSION

Analysis of variance

Analysis of variance for quantitative morphological traits analyzed for nineteen genotypes of chickpea revealed highly significant ($p < 0.01$) differences among genotypes for GY, HSW, BYD, PHT, DTF, NSP, NPP and HI; and significant ($p < 0.05$) differences for NSB and DTM and there were no significant ($p > 0.05$) differences for NPB (Table 2). This indicated that these traits were relatively sensitive to environmental effects and reflected insignificant variability. Also, there were highly significant ($p < 0.01$) differences of all traits for the blocks excepting NPB and NSB ($p > 0.05$). The mean values of all tested morphological traits of nineteen released chickpea varieties are presented (Table 1) showed significant differences between most of the studied traits. Based on the means of different released chickpea cultivars for the yield and various parameters (Table 3), high phenotypic variations were recorded among the nineteen released/elite Ethiopian chickpea varieties. From the results of this study, the variety ICCV-92069 had shown significantly less number of secondary branches than others. DZ-10-4 had shown significantly less number of primary branches than Minjar, ICCL-80106, Dubie, ICCV-92006, ICCV-93512, Mariye, ICCV-92318, ACOS Dubie, ICCV-14808, FLIP88-42C and FLIP89-84C. These were followed by chickpea genotypes ICCL82104, FLIP-97-63C, ICCV-92069, ICCX-910112-6, DZ-10-11 and FLIP-97-66C. Chickpea genotypes (ICCL-80106, ICCV-82033, ICCV-93512, ICCV-14808, ICCV-92069 and ICCX-910112-6) had significant difference in the biological yields per plant as compared to ACOS-Dubie, FLIP-97-66C and FLIP-97-63C. Genotype ICCV-14808 had significantly more number of seeds per plant than FLIP-97-66C, Dubie and ICCX-910112-6 chickpea cultivars (Table 4). Chickpea genotype ICCV-92069 had smaller plant height than FLIP-97-63C, ICCV-93512, FLIP-97-66C, ICCX-910112-6, ICCV-14808, Dubie, FLIP-88-42C, FLIP-89-84C, ICCL82104 and ICCL80106. In case of pods per plant, chickpea genotypes (Mariye, ICCV-14808, ICCV-92069, DZ-10-11 and ICCL-80106) had significantly more pods per plant than FLIP88-42C, Dubie, ICCX-910112-6 and FLIP-97-66C.

There were significant differences between ACOSDubie and FLIP-97-66C for harvesting index. Both varieties had

Table 2. Mean square for quantitative morphological traits of chickpea cultivars analysis of variance (ANOVA).

Source of variation	d.f	Mean squares										
		HSW	GY	BYD	PHT	NPB	DTF	NSB	NSP	NPP	HI	DTM
Replication	2	26.34**	266355.3**	658220.6**	11.28**	0.03	10.8**	0.60	71.94**	113.29**	190.1**	321.9**
Genotypes	18	64.2**	9230.9**	56393.8**	15.3**	0.24	15.2**	11.6*	208.0**	125.7**	94.9**	11.1*
Error		1.33	20.83	35.10	0.49	0.02	0.56	0.20	2.16	1.58	1.25	0.61
CV (%)		36.75	27.26	23.44	10.203	7.96	9.27	18.43	36.94	30.93	18.86	18.85
Mean		27.19	442.4	905.45	34.95	2.33	44.94	8.169	43.97	37.95	48.08	110.33

** and *, significant at 0.01 and 0.05 probability level, respectively. HSW, 100 Seed weight; BYD, Biological yield; GY, Grain yield; PHT, Plant height; DTF, Days to 50% flowering; NPB, Number of primary branches; NSB, Number of secondary branches; NSP, Number of seeds per plant; NPP, Number of pods per plant; HI, Harvest index; DTM, Days to 90% maturity.

significantly greater harvesting index than ICCV-93512 and ICCV-14808 cultivars. There were no significant differences being observed among the varieties for high hundred seed weights according to LSD test, which was seen for varieties such as ICCX-910112-6, Dubie, FLIP-97-63C and ICCV-93512. However, these genotypes had significantly greater hundred seed weight than all other genotypes. ICCV-93512 and ICCV-14808 genotypes take relatively longer days to mature than all other genotypes, except for Dubie which mature early than the other genotypes. Similarly, genotypes like ICCV-92006, FLIP89-84C, Mariye, ICCL82104 and ICCV-92069 relatively showed high grain yield per plant than all other genotypes. In addition, no significant variation was observed in the flowering days among all the varieties tested. Furthermore, the results suggested the presence of sufficient variability among genotypes for days to maturity, primary and secondary branches per plant, plant height, 100-seed weight, and grain yield per plant. This result is consistent with Rehman et al. (1996) who reported similar results for chickpea genotypes in biological yield, grain yield number of seeds per plant and harvest index. Feven (2002) and Melese (2005) also reported highly significant difference among populations for most of the traits such as days to maturity, grain yield per plant, biological yield per plant and harvesting index; indicating the scope for selection of various morphogenetic traits from these highly diversified genotypes. The report is supported by other authors who reported the presence of genotypic variability in chickpea; such as Kumar et al. (1999), Nimbalkar (2000), Wahid and Ahmad (1999) who observed significant variation for a number of seeds per pod and per plant, seeds per plant, hundred seed weight and yield per plant, respectively. Several chickpea investigations recorded significant genotypic differences among the crop collections studied by them (Chander et al., 2001; Abdalla et al., 2003; Zerihun et al., 2018).

Genotypic and phenotypic coefficient of variation

Mean, range and coefficient of variation of agronomic traits have been widely used to determine the variations available in the population. Moreover, the values of genotypic and phenotypic coefficient of variations, >20%, 10 to 20% and <10% are considered to be higher, intermediate and lower respectively (Getachew et al., 2015). The effectiveness of selection in any crop depends on the extent and nature of phenotypic and genotypic variability present in different agronomic traits found in the population (Arora, 1991; Keneni et al., 2011). For this study, lower coefficients of variation (1.20 to 6.27%) were found for number of primary branches per plant, days to maturity and number of secondary branches per plant; and the results are consistent with the findings of Muhammad et al. (2005). Moderate variations (10.95-19.85) were observed for days to 50% flowering, plant height, hundred seeds weight and harvesting index's per plant and higher (26.69 to 38.23) for grain yield per plant, number of pods per plant, number of seeds per plant and biological yield per plant (Table 4). Similar results were reported by Deressa et al. (2013) and Muhammad et al. (2005). The genotypic coefficients of variation were also found lower for biological yield (6.10%) and moderate for number of primary branches (18.10%); while higher coefficient of variations were observed for days to flowering, plant height, days to maturity, number of secondary branches, hundred seed weight, number of pods per plant, harvest index, number of seeds per plant and grain yield per

Table 3. Means of different chickpea genotypes for the yield and various quantitative traits.

Varieties	Traits										
	DTF	PHT	NPB	NSB	NPP	NSP	HSW	DTM	BYD	GY	HI
Minjar	40.7	33.9abc	3.2a	9.0cd	36.9ab	47.7ab	21.7	110.3a	833.3bcde	369.3	44.9abcd
ICCV-92318	46.0	34.6abc	2.4d	13.4a	38.3ab	40.4ab	28.9	107.7ab	866.7bcde	422.3	48.5abcd
DZ-10-4	46.3	33.7abc	2.0g	9.8bc	38.4ab	48.1ab	24.7	109.0ab	966.7abcd	422.7	43.7abcd
ICCV-92033	43.7	31.1bc	2.6c	13.4a	38.9ab	46.1ab	25.5	110.3ab	1100.0ab	439.7	39.7bcd
Acos Dubie	40.7	32.9abc	2.4d	9.6bc	37.4ab	46.7ab	29.8	107.7ab	600.0e	361.7	59.4a
ICCV-92006	44.7	34.3abc	2.6c	10.0b	34.3ab	37.3ab	30.0	109.7ab	966.7abcd	492.7	50.0abc
ICCL82104	47.7	35.3ab	2.2f	7.6fgh	33.8ab	40.2ab	26.3	109.7ab	983.3abcd	469.3	49.4abcd
FLIP-97-63C	45.3	38.1a	2.2f	7.4fgh	32.1ab	36.1ab	33.9	109.3ab	800.0cde	326.0	41.3bcd
ICCV-93512	45.3	37.2ab	2.6c	8.6de	32.1ab	35.0ab	30.1	114.0ab	1066.7abc	391.7	38.7cd
ICCV-14808	40.7	35.9ab	2.4d	6.4ij	44.8a	58.5a	21.3	112.3ab	1066.7abc	346.3	33.03d
Mariye	42.3	31.7bc	2.6c	8.2def	45.4a	49.9ab	22.7	109.0ab	900.0abcd	479.0	53.8abc
ICCL-80106	46.0	35.1ab	3.0b	8.2def	40.2a	48.7ab	22.9	109.7ab	1166.7a	491.3	43.4abcd
ICCV-92069	43.0	28.7c	2.2f	6.0j	43.6a	49.5ab	20.9	109.3ab	1033.3abc	463.7	44.97abcd
FLIP-97-66C	46.0	36.3ab	2.3e	7.3ghi	29.7ab	31.8b	27.5	111.0ab	733.3de	407.7	55.6ab
Dubie	41.0	35.9ab	2.43d	9.6bc	27.7ab	29.8b	34.1	106.0b	983.3abcd	417.7	41.8bcd
ICCX-910112-6	46.7	36.1ab	2.2f	6.8hij	28.1ab	29.9b	37.1	108.0ab	1033.3abc	445.3	42.6bcd
FLIP88-42C	46.3	35.5ab	2.4d	7.2ghi	20.4c	38.2ab	26.7	110.3ab	900.0abcd	382.3	42.4bcd
DZ-10-11	44.0	33.1abc	2.2f	8.0efg	40.9a	52.6ab	25.8	107.3ab	866.7bcde	399.0	48.4abcd
FLIP89-84C	45.3	35.47ab	2.4d	9.0cd	34.3ab	37.9ab	26.1	109.3ab	933.3abcd	372.7	39.6bcd
Mean	44.3	34.47	2.44	8.7	35.66	42.33	27.16	109.46	936.84	415.8	45.34
LSD	8.37^{NS}	6.27	0.05	0.90	19.21	25.34	17.7^{NS}	6.83	270.96	192.6^{NS}	16.62
CV %	10.95	10.984	1.20	6.273	31.54	34.95	36.75	3.77	17.46583	26.69	19.85

Means sharing the same letters are non-significant at the 0.05 and NS: non-significant ($p > 0.05$) probability levels, respectively according to Least Significant Difference (LSD) and CV = Coefficient of variation. HSW, 100 Seed weight; BYD, Biological yield; GY, Grain yield; PHT, Plant height; DTF, Days to 50% flowering; NPB, Number of primary branches; NSB, Number of secondary branches; NSP, Number of seeds per plant; NPP, Number of pods per plant; HI, Harvest index; DTM, Days to 90% maturity.

plant. Moderate genotypic coefficients of variation were observed for days to maturity and number of primary branches (12.73% and 16.90%, respectively). Higher genotypic coefficients of variation were observed for the remaining characters (>20%) (Table 4). Overall, no lower genetic coefficient of variability was recorded in this study. The recorded ranges for the quantitative traits indicated the presence of variation among chickpea genotypes. In this study, the range of quantitative traits for biological yield, number of pods per plant, grain yield, number of seeds per plant and hundred seeds weight showed the existence of considerable variation. However, days to maturity, number of primary and secondary branches had relatively low range that is indicated to be relatively low as compared to the other quantitative traits and the same result were also reported by Muhammad et al. (2005), and Pundir et al. (1991). Rao and Kumar (2000) and Singh et al. (1990) reported low variability for days to maturity, while moderately high phenotypic coefficients of variability was noted by Arora (1991) for primary branches per

Table 4. Mean, range, phenotypic variance, genotypic variance and environmental variance, phenotypic coefficient of variation and genotypic coefficient of variation of quantitative traits of the chickpea genotypes.

Characters	Mean	Range	CV	PV	GV	EV	PCV	GCV
Hundred seeds weight (gm)	27.16	15.63-55.73	17.47	17.94	1.91	16.03	81.23	26.50
Days of 50% flowering	44.30	38.33-51.67	10.95	3.67	3.44	0.23	28.81	27.89
Days of 90% maturity	109.46	104-116	3.77	25.14	1.77	23.37	47.96	12.73
Grain yield	415.81	252 -601.33	26.69	3039.98	251.25	2788.73	270.41	77.74
Harvest index	45.34	27.77-56.00	19.85	68.69	28.57	40.12	121.57	78.40
Biological yield	936.84	733.3-1166.7	38.23	18731.0	3.49	18727.54	46.72	6.10
Number of primary branches	2.44	2.27-2.73	1.20	0.08	0.07	0.01	18.10	16.90
Number of pods per plant	35.66	15.3-52.4	31.54	38.15	6.30	31.85	103.35	42.00
Number of secondary branches	8.69	6.8-10.6	6.27	4.52	3.70	0.82	72.12	65.25
Number of seeds per plant	42.33	15.3-71.27	34.95	64.40	15.09	49.31	123.33	59.70
Plant height (cm)	34.47	29.87-39.33	10.98	7.85	3.84	4.01	47.67	33.34

CV, Coefficient of Variation; PV, Phenotypic Variance; GV, Genotypic Variance; EV, Environmental Variance; PCV, Phenotypic Coefficient of Variation; GCV, Genotypic Coefficient of Variation.

Table 5. Principal component analysis of quantitative traits of chickpea genotypes.

Principal component	HSW	GY	BYD	PHT	NPB	DTF	NSB	NSP	NPP	HI	DTM
1	-0.47	0.35	0.23	-0.32	0.10	0.06	0.10	0.49	0.45	0.18	-0.07
2	0.17	0.46	0.55	0.34	0.00	0.47	0.19	-0.20	-0.16	0.15	-0.02
3	-0.15	-0.20	0.17	0.14	0.42	-0.04	0.48	0.06	0.02	-0.46	0.51
4	0.09	-0.12	0.10	0.21	0.25	-0.52	0.47	-0.02	-0.01	0.36	-0.48

HSW, 100 Seed weight; BYD, Biological yield; GY, Grain yield; PHT, Plant height; DTF, Days to 50% flowering; NPB, Number of primary branches; NSB, Number of secondary branches; NSP, Number of seeds per plant; NPP, Number of pods per plant; HI, Harvest index; DTM, Days to 90% maturity.

plant and 100-seed weight. Khan and Sharma (1999) reported high genetic coefficient of variation for secondary branches per plant. Rehman et al. (1996) and Wahid and Ahmed (1999) reported high estimate of genetic coefficient of variability for plant height and seeds per pod. Getachew et al. (2015) reported high genetic coefficients of variability for seeds per plant and seed yield per plant. High genotypic coefficient of variation indicated the availability of high genetic variation for selection and improvement; while the lower value indicated that selection is not effective for particular character because of the narrow genetic variability (Singh et al., 2003; Upadhaya et al., 2008; Mullualem et al., 2017; Shiferaw et al., 2017).

Correlation coefficients of quantitative traits

The associations among traits are useful for selection of genotypes possessing groups of desired characters. Grain yield per plant had highly significant and positive correlations with biological yield, days to 50% flowering, number of seeds per plant and number of pods per plant, and number of primary branches per plant. The yield components exhibited varying trends of association among themselves (Table 5). In contrast, days to 50% flowering showed negative and insignificant phenotypic correlations with primary branches, secondary branches, number of seeds per plant and hundred seeds weight. Days to 50% flowering showed positive but insignificant correlation with plant height and number of pods per plant.

Conclusions

The phenotypic and genotypic coefficients of variation range for quantitative traits and analysis of variance confirmed the existence of variability among released chickpea varieties. Similarly, coefficient of variation for quantitative morphological traits indicated the availability of variation within the same. In the present study, most of the traits had medium to high variation, implying that there is genetic variability among nineteen released chickpea varieties. The highly strong and positive significant correlation recorded between grain yield and

biological yield**,** days to flowering, number of seeds per plant and number of pods per plant. The positive significant correlation with number of primary branches per plant indicated that the yield components exhibited varying trends of association among themselves and the improvements of one trait will affect yield improvement of chickpea. Therefore, the morphological diversity analysis has shown that there is a considerable genetic diversity among the Ethiopian released chickpea varieties, which can be used for further improvement of the released varieties.

CONFLICT OF INTERESTS

The authors declare that they have no conflict of interest.

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Full Length Research Paper

Determination of optimum plant density of broadcasting, row planting and transplanting on yield and yield components of Tef [*Eragrostis tef* (Zucc.) Trotter] in Central Zone of Tigray, Ethiopia

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The low national tef productivity is mainly attributed by lack of improved agronomic practices. The row planting and transplanting is one of the promising planting techniques proved to boost the yield of tef. The objective was to determine the optimum seed rate, row, broadcasting and plant population in intra and inter row spacing of transplanted tef. A field experiment was carried out at Laelay maychew and Naeder adet for two years. The experiment was laid out in RCBD with three replications having 3×3 (9 m^2) and seven treatments. The analysis of variance showed that there were a significance difference at (P≤0.001) for days to maturity, plant height and panicle length, whereas grain yield was significantly difference at (P≤0.01). The treatment with transplanting 20 cm intra row spacing distance resulted in the highest grain yield of 2586 $kgha^{-1}$ followed by broadcasting at 5 $kgha^{-1}$ (2547.2 $kgha^{-1}$). Even though there was a significant difference among the treatments, combined mean performance of grain yield showed that there was no statically significant difference mainly among treatments of transplanting with 20×20 and 20×15 cm inters and intra row spacing and broadcasting at 5 kg ha-1 which was recorded as the higher grain yield. The present study recommended that the use of the broadcasting with seed rate of 5 $kgha^{-1}$ and even though the transplanting is a labor consumer, it might be important for early drought faced tef growing areas with intra row spacing of 20×15 cm depending on the rain fail condition. Further tef planter machine development for lower seed rate is needed.

Key words: Tef transplanting, seeding rate, row spacing.

INTRODUCTION

Tef [Eragrostis tef (Zucc.) Trotter], which has genetic origin and center diversity in Ethiopia (Vavilov, 1951). Tef is an important staple cereal crop in Ethiopia occupying more than three million hectare of land. It is first in area coverage but second and last in production and productivity, respectively, from cereals under production in Ethiopia. It is grown by over 6.6 million households and constitutes the major staple food grain for over 50 million Ethiopians (CSA, 2015).

Nutritionally, tef has been receiving global attention as health food because of its gluten-free nature that renders it suitable for people suffering from gluten allergy known

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as celiac disease and slow release carbohydrates that make it suitable for diabetic people. Antibody-based assay have shown that tef does not contain the offending epitopes (Spaenij-Dekking et al., 2005). Prossesion of the genomic sequence allow for confirmation of these assay (Cannarozzi et al., 2014). Tef has high iron content that makes it suitable for pregnancy-related and hookworm infestation related anemia (Alaunyte et al., 2012). The iron content seems to play a particularly important role in Ethiopia, as absence of anemia has been found to correlate with areas of tef consumption (BoSTID, 1996).

The current low yield levels can be attributed to different production constraints such as susceptibility to lodging, moisture stress, and poor pre- and post-harvest agronomic management practices (Abrha, 2016). It has been argued that more efficient agronomic management could double the yield of crop plants (Mueller et al., 2012). Currently, the majority of the farmers practise broadcast sowing, which is associated with a high incidence of lodging, reduced plant growth and yield (Asargew et al., 2014).

The low national or regional tef productivity is mainly attributed by lack of improved agronomic practices. Broadcast method of sowing has been predominantly used in the past years; however, new agronomic practices could increase the productivity of the crop. Row planting and transplanting method of a month age tef seedlings are one of the promising planting techniques to boost the yield of tef (Seyfu, 1997; Kebebew et al., 2011). Transplanting is assumed to have the benefits of escaping dry spells occurring in any particular season and enhancing productivity under dry land areas. Transplanting in a row considerably increased the seed yield compared to the broadcasting method. In addition to this, it reduces the seed rate compared with the broadcasting method that a farmer uses 25 to 50 kg/ha tef as compared to transplanting required only 2 to 2.5kg/ha (Tareke et al., 2013).

The main effect of transplanting is increasing tiller number, producing strong and fertile tiller culms, increasing the number of productive tillers, which increased number of seeds/panicle. Best results came from wider spacing, giving individual plants wider space to show their potential, and the use of complete fertilizers (Zewdie, 2010). Transplanting is commonly practiced as a method of weed control for wet soil. Since the seedlings are vigor than the weed will help for control. While, requiring less seed transplanting require much more labor as compared to direct seeding (unpublished). Therefore, the present study was conducted to determine the optimum seed rate and plant population in intra and inter row spacing of transplanted tef.

MATERIALS AND METHODS

The field experiment was carried out at Axum Agricultural Research Center (AxARC) in Laelay Maychew (vertisoil) and Naeder-adet (light soil) districts of the Central Zone of Tigray, Northern Ethiopia, during the main production season of (July-November) 2012 and 2013. The sites are located at 250 km and 284 North West of Mekelle and 1024 and 1069 km North of Addis Ababa at a latitude of 14°07' 235" and 13°06'762"N, at a longitude of 038°43'987'' and 038°48'38'' E, at an altitude of 2118 and 2121 m above sea level, respectively. The experiment was lay down by the design RCBD with three replications and plot size of 3 m length and 3 m width (9 m^2) and spaces between plot and replication was 1 and 1.5 m, respectively.

Variety Quncho was used as an experimental material with the different seed rates. The treatments were (1) broadcasting at 5 $kgha^{-1}$, (2) broadcasting at 25 $kgha^{-1}$, (3) 15 cm, and (4) 20 cm of inter row spacing at 5 kg ha^{-1} seed rate for row planting and (5)10, (6)15, and (7) 20 cm intra row spacing and 20 cm inter row spacing for transplanting methods. Both the broadcasting and row planting were planted by hand drilling left on the surface little bet compacted by labors. The inter row spacing for row planting method were adjusted as 15 and 20 cm whereas for transplanting 20 cm inter row and 10, 15 and 20 cm intra row spacing were used. The treatment combinations are structured in Table 1.

According to the recommendation, tef fertilizer application for black soil 60 kg ha^{-1} P_2O_5 and N at Laelay maychew (Hatsebo) was applied and 60 kg ha^{-1} P_2O_5 and 40 kg ha^{-1} N also for light soil at Naeder–adet was applied (Seyfu, 1997). DAP was applied at planting and urea was applied in two splits, half at the time of planting and the remaining half at tillering stage. Seedling for transplanting was grown in a bed and transplanted to experimental plot at one month age (about three to four leaf atage), with three seedlings per hill at the spacing per the treatment.

The time of transplanting was in the morning for better survival of the seedling. The experimental materials were sown on the second week of July 2012 and 2013 main production seasons. All other pre and post-planting management practices were done in accordance with the research recommendations for tef production in the area.

Days to maturity were determined from 50% seedling emergence to 90% physiological maturity. Plant height in centimeters was measured from the base of the plant to the tip of the panicle on the primary tiller of five randomly selected plants per plot. Panicle length of the central tillers in centimeters was measured as the average length of the panicle from the node where the first panicle branch starts to the tip of the central tiller of five randomly selected plants per plot. Whereas, the grain yield (kg ha^{-1}) weighed the grain harvested from entire plot and the average was used to statistical analysis. Data were collected on plant and plot based and analyzed by SAS software version 9.1.3 (SAS Institute Inc. 2004) to evaluate the variance and mean separation using LSD at alpha level $\alpha \leq 0.05$.

RESULTS AND DISCUSSION

The analysis of variance (ANOVA) shows there was a statistically significant difference at ($P \leq 0.001$) for days to maturity, plant height and panicle length, whereas grain yield was significantly different at ($P \leq 0.01$). The highest grain yield was recorded for transplanting in 20×20 cm row spacing (2586.5 kg ha^{-1}) followed by broadcasting at 5 kg ha^{-1} (2547.2 kg ha^{-1}), transplanting (20×15 cm) at 2456.2 kg ha^{-1} and the row planting at 5 $kgha^{-1}$ (20 cm, 2279.00 $kgha^{-1}$) (Table 1). Transplanting was greater than both direct planting methods. The present result was similar to Tareke et al. (2013) who reported that transplanting had the highest grain yield than broadcasting and row planting. Fekremariam et al. (2014) reported that transplanting tef gave a yield advantage ranging from 29.2 to 39.3% over broadcasting

Table 1. The combined mean performance of tef yield and yield components evaluated by the different planting methods, seed rate, inter and intra broadcasting, row planting and transplanting at Laelay Maychew and Naeder Adet in 2012 and 2013 cropping season.

Treatment	DM (days)	pH (cm)	Pan (cm)	Gy (kg ha^{-1})
Broadcasting at 5 kg/ha	113.92^{a}	122.18^{ab}	49.63^{ab}	2547.20^{a}
Broadcasting at 25 kg/ha	95.92^{b}	104.91^{c}	43.63^{c}	1719.70^{b}
Row planting (20 cm) at 5 kg/ha	115.08^{a}	122.97^{ab}	49.88^{ab}	2279.00^{a}
Row planting (15 cm) at 5 kg/ha	97.17^{b}	108.51^{c}	43.23^{c}	1723.90^{b}
Transplanting (20×10 cm)	97.33^{b}	113.23^{bc}	46.63^{bc}	2134.20^{ab}
Transplanting (20×15 cm)	97.00^{b}	123.54^{ab}	51.45^{a}	2456.20^{a}
Transplanting (20×20 cm)	113.58^{a}	131.40^{a}	53.98^{a}	2586.50^{a}
Grand mean	104.28	118.10	48.35	2206.65
Coefficient of variance	9.93	12.60	11.35	33.11
Least significance difference	8.44	12.14	2.39	595.92
R-square	0.64	0.56	0.61	0.42
Treatments × locations(α≤0.05)	0.3103	0.0768	0.0080**	0.5597^{ns}
Treatments(α≤0.05)	<0.0001**	0.0004**	<.0001**	0.0118*
Location(α≤0.05)	<.0001**	<.0001**	0.0035**	0.0002**

DM: Days to maturity, pH: plant height, Pan: panicle length, Gy: grain yield, * and ** significance , p<0.05 and p<0.01, respectively.

method. Even though there was a significant difference among the treatments the combined mean performance of grain yield showed that there was no statistically significant difference between treatments mainly between broadcasting at 5 kg ha^{-1}, row planting at 5 kg ha^{-1} with 20 cm and transplanting with 20×20 cm and 20×15 cm inter and intra row spacing, which was recorded the highest grain yield. The highest grain yield was recorded with wider inter and intra row spacing aligned with previous report (Zewdie, 2010).

For optimum and well distribution of rainfall, both broadcasting and row planting with seeding rate of 5 kgha^{-1} might enhance yield. Whereas, the transplanting method economically important when the environment is faced with early drought. Since, it is time consuming and labor intensive, Abraha et al. (2016) reported that transplanting maximized the yield of tef, but a cost-benefit analysis showed that row sowing was more profitable. Practically, the lower seed rate on small scale farming had a draw back in establishing of seedling with erratic rain fail in Tigray region; unless, there is development of tef planter. The results depend on the condition that low seed rate and transplanting increase the productivity of tef.

Days to maturity had a significant difference among the treatments. Broadcasting and row planting at 5 kg ha^{-1} of seed rate at 115 and 113 days, respectively and for the transplanting at 113 days scored longer days to maturity. Whereas, the shorter days to maturity were observed at the higher seed rate of broadcasting at 25 kgha^{-1} for 96 days and narrow inter and intra row spacing and transplanting at 5 kg ha^{-1}. This could be because of competition due to high population for nutrient and moisture was limited. The reason for the longest days to maturity might be the lower seed rate or population had slow growth due to the less competition among the individual plants. Therefore, this competition leads to late maturity. In addition to this, the short plant height was recorded from the higher seed rate of broadcasting at 25 kg ha^{-1} and 15 cm inter row spacing at 5 kg ha^{-1} of seed rate. The longest plant height was obtained from the transplanting 20×20 cm. Meantime, the longest panicle length was measured from the transplanting 20×20 cm, the best determined population density for production and productivity of tef.

CONCLUSION AND RECOMMENDATION

From the examined treatments from the two locations and two seasons, with broadcasting, row planting and transplanting type of planting methods, the highest grain yield was obtained from transplanting 20 × 20 cm (2586.5 kgha^{-1}) followed by broadcasting at 5 kgha^{-1} (2547.2 kgha^{-1}) and transplanting 20 × 15 cm (2456.2 kgha^{-1}) followed by row planting at 5 kgha^{-1} (2279.00 kgha^{-1}). The present study recommended that use of row planting at 5 kgha^{-1} and broadcasting with seed rate of at 5 kgha^{-1}, and even though transplanting is labor intensive it might be important for early drought growing areas of tef with intra row spacing of 20 × 20 cm. However, the low seed rate had a problem of tef seedling establishment at large farm field. Unless there is an appropriate tef planter and field leveling machine. The reason for the low seedling stand establishment is due to unevenly distribution of seeds during sowing. Thus, use of low seed rate is if a manually or motor-driven broadcaster or drill in available for both large and small scale farmers in the study areas and

similar agro-ecology.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests

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Generation and evaluation of heterogeneous genotypes of tomato for small-scale farmers

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Tomato is broadly distributed in tropical and subtropical America, where small farmers cultivate commercial and landraces or heirloom genotypes, which exchange genes within them when are planted in the same plot. In this context, three different genetic groups of tomato were evaluated for agromorphological and yield traits under greenhouse to assess the differences in function of the genotypic homogeneity and heterogeneity. Twenty-four non-conventional hybrids (F_1, population-x-advanced lines), seventeen landraces and six advanced lines (F_8) were evaluated in a randomized complete block design with three repetitions. Significant differences ($p \leq 0.05$) were determined among genetic groups for all variables evaluated, except in days to ripening of fruits at the fifth branch, and within genetic groups, significant differences were also detected. Six hybrids, three landraces and two advanced lines presented remarkable agronomic responses in yield per plant. The hybrids and landraces had high phenotypic variability in plant and fruit traits, with flat-rounded or lightly flattened fruit shapes, qualities demanded in the local markets, and a yield of 2 kg per plant. In Oaxaca, Mexico, small-scale farmers readily accept these heterogeneous genetic groups of tomato. High homogeneity characterized the advanced lines, with a fruit shape convenient for national and international markets.

Key words: Landraces, non-conventional hybrids, phenotypic divergences, principal component analysis.

INTRODUCTION

The tomato (*Solanum lycopersicum* L.) is an important economic and social horticultural crop, and the cultivation of tomato promotes a dynamic economy and generates employment in exporting countries. In the last decade, approximately 4.7 million ha are annually planted to tomato (FAOSTAT, 2014), with a consequent worldwide demand for seed of improved varieties every year. However, at the country level, different production systems are in operation, and the delivered varieties are not stable over all environments and greenhouse conditions. Therefore, each country that produces tomato must develop strategies to solve the problem of access

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for farmers to improved varieties, who demand at least two varietal groups; first, varieties for the export market and second, seeds for varieties for the national market. Unit size exploited and whether a conventional or organic production system must also be considered. To access genetic material of tomato, breeding programs have generated varieties with different genetic structure, including open-pollinated (OP) and synthetic varieties (SV), hybrids from triple (TH), double (DH) and simple (SH) crossings, non-conventional hybrids (that is, bred lines-x-OP or SV, OP-x-SV, SV/OP-x-landraces, and landrace-x-landrace, among others) and interspecific hybrids generated by cultivated and wild species using isogenic lines.

In the genetic improvement of tomato, increased yield and environmental stability across production systems are criteria used for selection. More recently, improved nutritional quality of fruit and a long shelf life were added as indispensable criteria. However, the task is complex, not simple, to join yield and nutritional-nutraceutical attributes in a variety. Traditionally, breeders use assistance from molecular markers and biochemical analysis of fruit quality to generate hybrids and synthetic or open-pollinated varieties; however, these approaches are insufficient to meet the demand for varieties (Grandillo et al., 1999, 2011). In different countries, old varieties or heirlooms of tomato are being selected and conserved by farmers, and although these farmer varieties have phenotypic heterogeneity, the fruit quality is highly preferred by consumers. Some examples of such farmer varieties are 'Valenciano', 'Muchamiel' and 'De Penjar' in Spain (Cebolla-Cornejo et al., 2013); 'Pomodoro di Mercatello' (Rocchi et al., 2016), 'A pera Abruzzese' (Mazzucato et al., 2010), 'Pomodoro di Sorrento', 'Belmonte', and 'Canestrino di Lucca' in Italy (Parisi et al., 2016); a dozen heirlooms in Brazil (Vargas et al., 2015); 'Tomataki Santorinis' from Santorini Island of Greece (Koutsika-Sotiriou et al., 2016); and different local varieties from Eritrea in Africa (Asgedom et al., 2011). In México, tomato landraces are commonly found from north to south, in addition to ruderal forms of S. *lycopersicum* var. *cerasiforme* (Bonilla-Barrientos et al., 2014; Chávez et al., 2011; Sanjuan et al., 2014).

For export to the international market, the producer requires hybrids that are genetically homogeneous heterozygotes producing fruit of high commercial quality. By contrast, small-scale farmers require seeds of varieties with broad adaptability to heterogeneous production systems, such as those of heterogeneous homozygotes or heterogeneous heterozygotes, but also with fruits highly preferred by regional consumers who will pay a premium price for fruit quality. This type of farmer can avoid the high cost of obtaining hybrid seed, because the farmers require only a small quantity of seed that they can reproduce themselves (Bonilla-Barrientos et al., 2014; Cebolla-Cornejo et al., 2013; Mazzucato et al., 2010; Parisi et al., 2016). Farmers know that the varieties, agroecological conditions and crop management determine the flavor, taste and nutritional quality of the tomato fruit (Cebolla-Cornejo et al., 2011). Moreover, the product destination is local or organic markets in which the quality of fruit is more important than the yield per area (Rocchi et al., 2016). Koutsika-Sotiriou et al. (2016) compared breeding between farmers and formal plant breeding using tomato heirlooms and concluded that farmers generated populations with low productivity, high fruit homogeneity and broad adaptability, whereas the breeders produced advanced lines or selected populations with high productivity, high selection efficiency and specific adaptations. Therefore, farmers start selecting their varieties only to give them gene pools with broad genetic variability, which can help to maintain independence in access to seed without intervention of seed companies.

In recent decades, the organic markets for tomato and ecological agriculture have required seeds of varieties with high tolerance or resistance to pests, diseases and abiotic stresses but also with high quality fruit based on physical and chemical aspects. To develop such varieties, breeders resort to primary pools (advanced lines from plant breeding programs), secondary pools (farmer varieties, landraces or gene banks) and tertiary genetic material such as wild species or wild relatives of the cultivated species (Lammert Van Bueren et al., 2011; Riahi et al., 2009). In this context, the aims in this work were to evaluate a collection of tomatoes composed of non-conventional hybrids of simple crossings, farmer varieties and advanced bred lines under greenhouse conditions in a local system of low input agriculture to assess the productivity of heterogeneous genetic material in developing an agronomic proposal for small-scale farmers.

MATERIALS AND METHODS

Germplasms evaluated

The tomato collection included 47 genotypes from three different genetic groups: 24 non-conventional hybrids, F_1 from population-x-advanced line crosses (H-60, H-61, H-62, H-63, H-64, H-65, H-66, H-67, H-68, H-69, H-70, H-71, H-72, H-73, H-74, H-75, H-01, H-06, H-06a, H-12, H-12a, H-19, H-22 and H-22a); 17 samples of landraces from Oaxaca, Mexico (COMP 5, X-04, X-05, X-07, X-08, X-09, X-12, X-13, X-15, I-18, I-07, I-25, I-31, I-35, I-38, I-42 and I-51); and five advanced inbred lines (the F_8, LA-106, LA-107, LA-108, LA-110 and LA-112). In the first two groups, fruits are broadly variable in size and shape, including rounded, pyriform, flattened, slightly flattened, heart-shaped and other similar shapes, with shoulders or amorphous protuberances but with three or more locules. Locally, these groups are called 'criollo' or 'costilla' in Spanish.

Experiment management

The tomato collection was transplanted (August 4, 2015) in a complete randomized block design with three replications in a greenhouse (17° 01' 10.42'' N, 96° 45' 52.32'' W, 1561 m.a.s.l. and

Table 1. Significance of the mean square from the analysis of variance of evaluated traits in non-conventional hybrids, landraces and advanced lines of tomato.

Agromorphological variable	Genetic groups	Genotypes (groups)††	Repetition	Plant (rep.)††	CV (%)
Days of transplant to flowering of the 5th branch	132.8**	15.2**	3.6^{ns}	-	3.8
Days of transplant to setting of fruits in the 5th branch	292.32**	13.04**	17.58^{ns}	-	3.3
Days of transplant to ripening of the 5th fruit branches	18.87^{ns}	48.21**	24.31^{ns}	-	3.2
Plant height at 60 days after transplanting	21540.6*	9550.6*	10730.7^{ns}	5918.4^{ns}	4.8
Plant height at 90 days after transplanting	3940.1**	8629.6**	2148.1*	296.3^{ns}	9.5
Polar diameter (length) of fruit†	1536.1**	1828.3**	194.79*	25.76^{ns}	9.7
Equatorial diameter (width) of fruit†	156.01*	322.25**	35.99^{ns}	63.51^{ns}	10.8
Total number of flowers†	12555.2**	704.8**	314.95*	117.9^{ns}	21.0
Total number of fruits†	8612.1**	471.9**	180.4*	29.4^{ns}	17.0
Average weight of fruit	18184**	4283.7**	123.18^{ns}	178.5^{ns}	26.8
Total weight of fruits per plant†	6901444**	4439179**	718958^{ns}	204807^{ns}	17.1

^{ns}Not significant ($p > 0.05$); *significant at $p \leq 0.05$; **significant at $p \leq 0.01$; †variables evaluated at fifth floral and fruit branches; ††effect of genotypes nested in genetic groups of tomato and plants nested in repetitions; CV = coefficient of variation.

21.1°C exterior temperature, Oaxaca, Mexico). Before transplanting, the soil was removed to incorporate sawdust, cattle manure, lime and water, and at transplant, soils were treated with Captan®. During the cultivation, pruning, tutoring and staking of plants were the common practices, together with drip-fertilization using commercial formulas of 15-30-15, 18-18-18, and 13-6-40 (N-P-K) and calcium nitrate. Additionally, a preventive program of pest and disease management was implemented by applying preventive chemical products and vegetable extracts.

The agronomic behavior of the genotypes was evaluated throughout the study with physiological, morphological and agronomic variables. For example, in the experimental plots, the precocity was assessed with counts of days after transplant to reach flowering, fruit set and maturing fruit stages in 50% or more of plants at the level of the fifth floral branch. To determine growth habits, plant growth was evaluated with measurements of plant height at 60 and 90 days after transplanting. The primary traits associated with yield were total number of flowers and fruits per plant at the fifth floral branch, polar and equatorial fruit diameters, average fruit weight and yield per plant.

Statistical analyses

Different analyses of variance were performed on the database per experimental plot and genotype using a linear model of completely randomized blocks with nesting of tomato genotypes or populations into genetic groups of evaluation and for some response variables, nesting of number of evaluated plants in a genotype. All analyses of variance evaluated the differences among and within genetic groups, with the analyses complemented with multiple Tukey's tests ($p \leq 0.05$). Additionally, for the average per genotype for each variable, later standardized, two principal component analysis were conducted using a variance-covariance matrix to describe and assess the variables of high descriptive value in the agronomic behavior of the evaluated genotypes and its relationships with plant yield. All analyses were conducted in the SAS statistical software package (SAS, 1999).

RESULTS

Significant differences ($p \leq 0.05$) were detected among and within genetic groups of tomato for all variables, except in days to fruit ripening at the fifth floral branch (Table 1). The results showed different responses among genetic groups under greenhouse conditions, and in such responses, the genetic variability contained in the genetic groups was clearly a buffer mechanism or for resilience.

In the comparison of means among genetic groups, the advanced lines presented high homogeneity in days to flowering and fruit setting, compared with the non-conventional hybrids and landraces (Table 2). Particularly, the hybrids showed precocity in reaching flowering, fruit setting and ripening. The commercial maturation of fruits from the fifth branch approached 108 days after transplanting. Consequently, the first harvests were performed between 32 and 42 days after transplanting, which included the first floral branches. Therefore, before the harvest of the fifth branch, two or three harvests with high quality fruit have been conducted.

In advanced lines, the number of flower and fruits per branch was higher than that in hybrids and farmer landraces. Additionally, in the tomato landraces, the fruit setting was lower than that in hybrids and advanced lines, and only one-third of the total of flowers produced fruits. Nevertheless, these fruits were large in size and weighed approximately 200 g or more per fruit, which is a characteristic that is very attractive to small-scale farmers. The advanced lines averaged 3 kg of fruit per plant, which was higher than that in hybrids and landraces (Table 2), because such lines were in a selection process for eight cycles with selection by the bulk population method (Acqaah, 2012). Into each genetic group, all agromorphological traits were highly variable. For example, among population hybrids, the flowering of the fifth floral branch occurred between 49 and 59 days after transplant (dat), plant height varied from 1.9 to 3.0 m at 90 dat and fruits shapes were round,

Table 2. Comparisons of physiological and agronomic behaviors among three gene pools of tomato.

Traits	Non-conventional hybrids	Landraces	Advanced lines
Days of transplant to flowering of the 5th floral branch	53.3±2.4[c*]	55.4±2.2[b]	57.1±1.6[a]
Days of transplant to setting of fruits in the 5th branch	61.6±2.4[b]	65.7±1.6[a]	65.4±1.6[a]
Days of transplant to ripening of the 5th fruit branches	107.7±4.6[a]	108.1±3.1[a]	109.3±3.5[a]
Plant height at 60 days after transplanting (cm)	179.2±19.6[a]	162.8±12.0[b]	159.0±27.7[b]
Plant height at 90 days after transplanting (cm)	237.0±26.2[a]	224.3±18.1[b]	212.3±35.8[c]
Polar diameter (length) of fruit† (mm)	56.5±12.5[b]	45.6±5.7[c]	61.0±9.6[a]
Equatorial diameter (width) of fruit† (mm)	54.8±5.1[b]	67.8±10.5[a]	49.7±4.2[c]
Total number of flowers†	39.6±5.6[b]	53.2±10.7[a]	38.0±4.3[b]
Total number of fruits†	23.4±6.0[b]	13.1±6.3[c]	27.9±7.5[a]
Average weight of fruit (g)	73.0±21.4[b]	83.9±37.9[a]	73.5±12.8[b]
Total weight of fruits per plant† (g)	1703.8±622.5[b]	1053.5±600.7[c]	2114.5±791.6[a]

*In row, means with same letter are not significantly different (Tukey's test, $p \leq 0.05$).

pyriform, saladette-type, round-flattened with shoulders and other shapes. The hybrids H-06, H-06a, H-22a, H-67, H-68 and H-72 had fruit set of more than 70%, measured by the relation fruits/flowers on the fifth floral branch. Seven non-conventional hybrids produced between 2.03 and 3.0 kg per plant (Table 3).

In the regional landraces, fructifying of the fifth branch occurred from 63 to 68 dat, plant height was from 1.9 to 2.4 m at 90 dat and the growth, which never stopped during the entire experiment, was considered indeterminate. These landraces had regularly round-flattened fruits with shoulders and the fruit set rate (fruits/flower) was low at 50%; only in the genotype I-25, fruit set reached 57.4%. Therefore, although these genotypes were highly variable in traits of plants and fruit shapes, variability in fruit setting was low. For landraces from Oaxaca, Mexico, the resulting lower yields were compensated with flavor, aroma and texture of fruit. In these cases, the yield varied from 0.27 to 2.06 kg per plant with an average weight from 52.0 to 192.6 g per fruit (Table 3).

The fruit of all advanced lines was saladette-type, and consequently, the length was major than equatorial diameter of fruit, except in the genotype LA-113a, which had a round shape. In these genotypes, the fruit setting rates (fruits/flowers) were from 75.5 to 88.7%, except in LA-106, with a rate less than 42%. A group of five lines presented high fruit weights (39.4 to 84.5 g) and uniformity in fruit shape of the commercial-type. Specifically, line LA-108 presented pyriform-enlarged fruits (7.7 cm in length) and line LA-113a had round-heart fruits, with yields up to 2.8 kg per plant for both lines (Table 3).

In the principal component analysis by morphological and physiological traits, the first principal component described 94.9% of total phenotypic variation, with eigenvalues of 0.58 and 0.81 for the variables plant height at 60 and 90 days after transplanting, respectively, and the physiological traits of flowering, fruit set and ripening of fruits with significant descriptive value (Figure 1). Based on yield traits, a second principal component (PC) analysis was performed, and in this case, the first component (PC1) described 71.5% of the total phenotypic variation, which was considered a discriminant index (PC1) of genotypic productivity. In this analysis, the variables of primary descriptive value for the total variability were polar (0.11 eigenvector) and equatorial (0.29 eigenvector) diameters and average weight of fruit (0.94 eigenvector). The relationships between yield per plant and first principal component or yield component index are shown in Figure 2. The landraces I-07, I-18 and I-31 had the highest values of equatorial diameter of fruit and high yield per plant. Similarly, the advanced lines LA-113 and LA-108 and the non-conventional hybrids H-06, H-06a and H-01 were outstanding within their heterogeneous genetic group.

DISCUSSION

Table 1 shows significant differences among genotype groups mainly due to high variability in each group, it was notorious that the advanced lines showed less variability than non-conventional hybrids and landraces. For plant height, the hybrids grew taller than the landraces (Table 2). Therefore, although the hybrids were non-conventional (crossing of lines x landraces or populations), the plants exhibited a hybrid vigor as result of the heterotic effect caused by genetic divergences among crossed parents. Mendoza-de Jesús et al. (2010) and Pinacho-Hernández et al. (2011) also observed heterotic effects in inter-population and inter-varietal crosses, respectively. These findings suggest that it is not only possible to exploit the hybrid vigor of the crossing of inbred lines but also that of crossing among populations or landraces-x-advanced lines, such as in this study. For both cases, lines or genetic populations without recent matching are the principle.

Table 3. Means comparison among populations within each tomato gene pool (ID, genotype H = non-conventional hybrids; X, I or COMP = landraces; LA = advanced lines).

Genotypes	DFL[1]	DFR	DMF	AP60	AP90	DPF	DEF	NFL	NFR	PMFR	RPP
H-01	54.0	63.7	107.7	172.9	223.3	54.6	58.5	32.5	14.5	104.5	1564.2
H-06	50.0	58.7	108.3	199.9	262.9	80.9	68.0	35.8	26.3	117.9	3003.8
H-06a	51.7	61.3	100.7	208.7	252.5	91.3	52.6	26.2	21.2	127.9	2596.0
H-12	56.3	64.3	110.0	165.2	229.6	55.0	55.6	35.4	22.7	73.8	1684.4
H-12a	56.3	64.3	107.3	179.4	235.0	51.2	55.8	38.7	27.7	72.3	1976.7
H-19	54.0	63.0	104.7	217.6	301.7	62.0	51.3	54.7	24.5	88.9	2042.4
H-22	51.0	61.3	103.7	163.7	218.3	45.5	50.6	41.9	23.9	62.1	1486.9
H-22a	54.0	60.3	110.0	154.7	216.2	46.4	53.0	40.8	26.7	63.6	1712.8
H-60	51.3	62.0	114.7	162.4	223.3	45.6	54.3	50.1	26.7	51.3	1365.0
H-61	57.7	66.3	116.3	145.3	193.7	54.5	46.2	40.8	11.6	47.1	581.3
H-62	56.0	66.3	114.3	140.6	195.4	54.1	48.5	39.7	19.5	52.3	1029.2
H-63	55.0	62.0	110.0	161.5	223.7	41.1	46.6	37.4	15.2	44.7	691.8
H-64	53.7	59.7	110.0	167.5	225.8	74.4	53.3	38.6	27.8	71.9	2025.2
H-65	53.0	61.7	111.7	183.2	256.2	56.9	58.5	37.4	15.7	49.8	780.3
H-66	58.7	65.3	114.0	165.4	236.7	49.9	52.0	44.3	11.4	66.2	709.8
H-67	52.3	59.0	109.0	176.3	251.7	56.1	55.7	38.6	29.5	86.2	2466.8
H-68	52.0	59.0	104.7	164.0	237.1	60.3	65.3	39.8	31.4	77.6	2437.8
H-69	49.3	59.0	100.0	195.2	268.6	43.6	57.3	45.7	26.5	82.4	2146.3
H-70	51.7	59.7	102.0	164.0	221.2	49.6	50.5	37.0	33.2	53.5	1769.8
H-71	52.0	62.0	103.3	150.5	212.5	74.1	57.0	39.9	24.5	68.3	1686.4
H-72	52.7	59.3	104.7	190.2	265.8	51.6	54.5	37.5	27.2	65.2	1776.5
H-73	53.3	61.0	108.3	159.0	208.7	53.6	58.9	34.8	22.7	89.4	1905.1
H-74	51.0	59.7	108.0	179.2	257.1	53.1	55.3	40.0	25.0	67.6	1682.0
H-75	51.7	58.7	101.0	191.3	270.0	49.9	55.3	41.8	26.6	67.8	1770.9
COMP5	55.7	66.7	110.7	168.7	232.9	41.2	73.5	57.2	13.3	86.7	1135.5
I-18	57.7	64.7	111.0	174.6	230.4	54.7	78.7	45.2	12.4	157.5	1880.6
I-07	59.0	65.7	113.3	132.2	187.8	52.3	97.1	38.0	9.7	192.6	1772.3
I-25	54.3	65.3	105.0	156.2	214.6	52.6	63.0	45.1	25.9	73.1	1520.8
I-31	57.3	63.7	111.0	152.3	197.9	45.6	70.8	44.9	17.5	119.6	2059.8
I-35	50.0	64.7	103.0	175.2	235.4	49.8	58.3	47.8	25.2	69.3	1639.3
I-38	56.3	64.7	110.0	164.3	203.3	52.3	63.4	43.5	20.1	82.8	1619.8
I-42	55.7	63.0	110.3	148.6	201.7	49.8	55.9	40.0	14.7	91.6	1131.9
I-51	55.0	65.3	107.7	150.0	207.9	45.0	61.0	40.0	18.1	72.7	1267.4
X-04	54.7	68.0	111.7	170.4	240.8	42.8	57.7	63.0	8.4	61.4	474.0
X-05	56.3	68.0	108.3	168.7	235.2	37.1	61.3	52.7	5.5	52.0	265.7
X-07	52.3	65.0	105.3	164.4	227.9	40.1	67.8	58.5	8.7	66.9	567.1
X-08	56.3	68.3	105.3	162.1	229.6	41.2	59.9	64.1	6.4	67.3	404.8
X-09	54.3	65.0	105.7	169.7	241.7	43.2	67.2	73.3	10.2	64.6	693.8
X-12	53.7	64.7	103.7	177.5	252.9	38.3	64.5	59.2	10.1	55.4	511.0
X-13	58.3	68.0	108.0	171.2	234.6	49.9	81.6	64.2	8.6	79.9	705.3
X-15	54.7	66.7	109.0	153.2	228.7	40.0	63.7	63.3	7.0	57.5	375.0
LA-106	59.7	68.0	115.0	117.7	152.9	56.2	46.0	35.2	14.5	49.9	734.6
LA-107	58.3	66.0	112.3	149.3	201.7	77.2	46.3	34.9	26.7	69.4	1816.2
LA-108	56.7	65.7	106.7	165.7	232.9	52.8	51.4	38.0	33.7	84.5	2828.5
LA-110	56.0	63.3	107.7	145.4	198.7	61.2	52.0	33.5	25.7	79.1	2065.4
LA-113a	56.0	64.0	106.7	180.0	234.2	52.0	56.3	42.1	34.7	83.0	2860.5
LA-113b	55.7	65.3	107.3	195.9	253.3	66.4	46.2	44.3	31.8	75.1	2381.8
DSH-Tukey	*7.9*	*7.9*	*13.1*	*28.7*	*37.1*	*9.8*	*16.1*	*16.8*	*11.2*	*44.5*	*915.0*

[1]DFL, days of transplant to flowering of the 5th branch; DFR, days of transplant to setting of fruits in the 5th branch; DMF, days of transplant to ripening of the 5th fruit branches; AP60, plant height at 60 days after transplanting (cm); AP90, plant height at 90 days after transplanting (cm); DPF, polar diameter (length) of fruit (mm); DEF, equatorial diameter (width) of fruit (mm), NFR, total number of flowers; NFR, total number of fruits; PMF, average weight per fruit; RPP, total weight of fruits per plant.

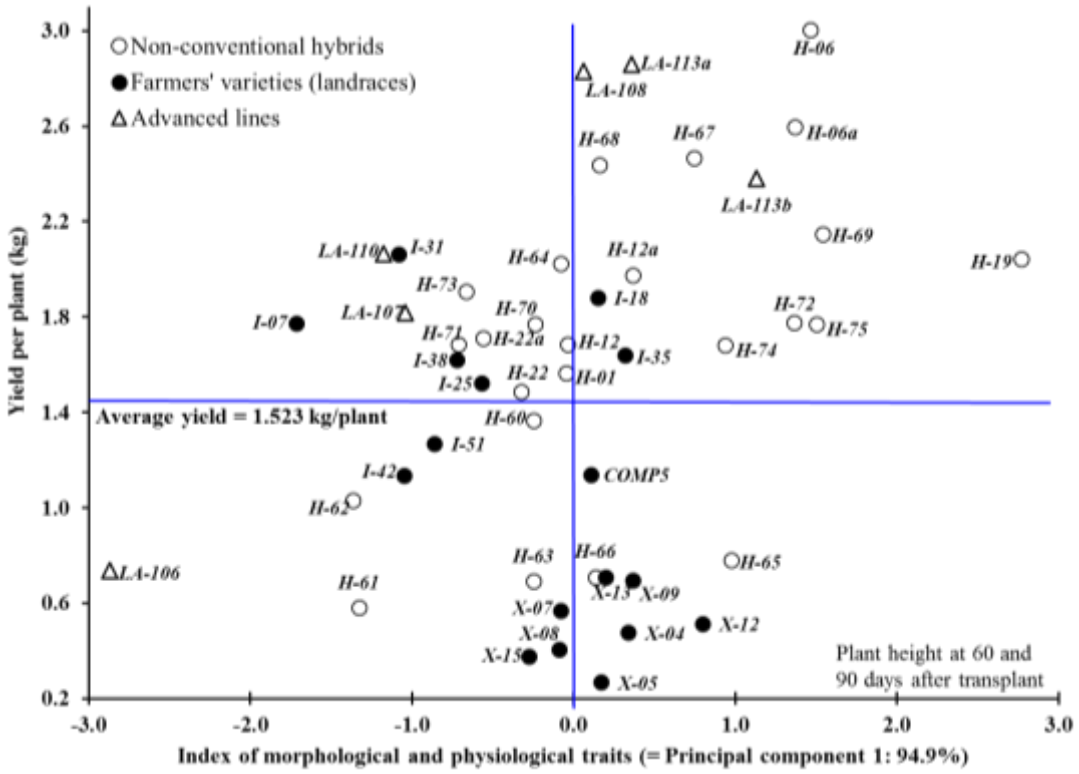

Figure 1. Relationship between plant yield and first principal component (index), based on morphological and physiological traits of plants.

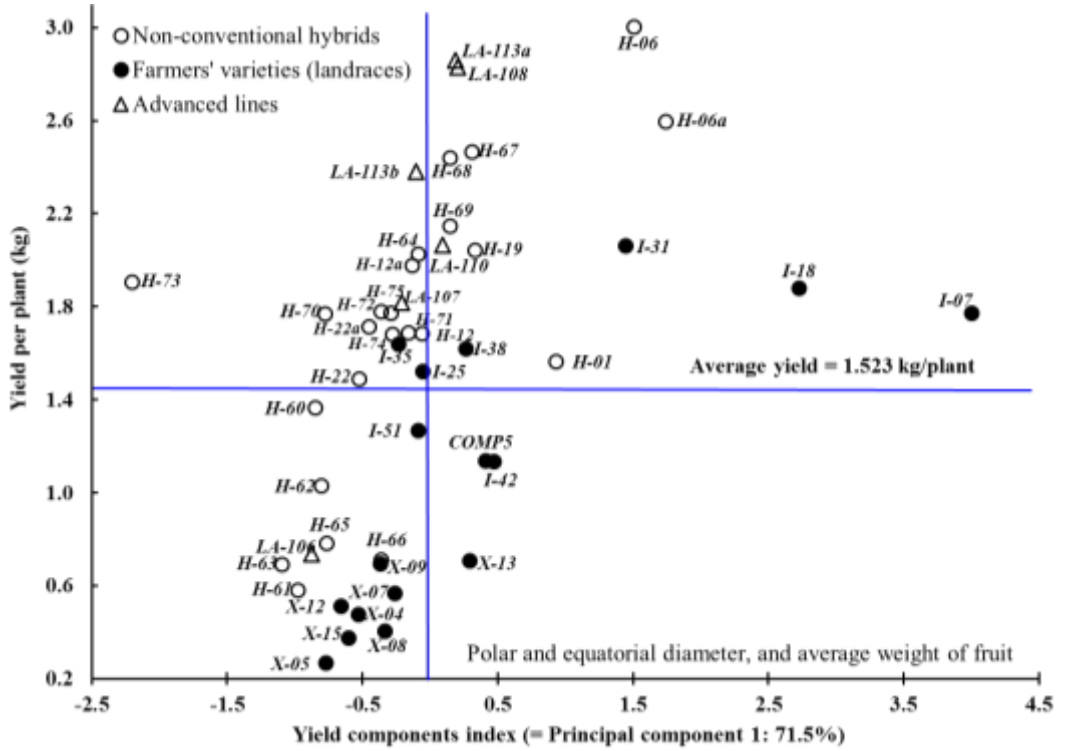

Figure 2. Pattern of relationships between yield per plant and first principal component (index), based on characters of yield.

In the size, shape and dimensions of fruit, the genetic groups showed phenotypic divergences in agronomic traits. For example, the fruits of advanced lines were oblong-elongated and lengthy; those of landraces were commonly rounded-flattened, lightly flattened or similarly shaped with shoulders and with a wide equatorial diameter; and those of non-conventional hybrids showed more variability of shape from rounded, saladette-type or round-flattened and high variation in size (Table 2). Phenotypic variation of these genetic groups offers opportunities for small-scale producers of tomato because of the necessity to diversify their production systems, which includes the production of fruit types for local or regional markets (specialties) and fruit shapes for the national market. Although the characteristics of plants and fruits may satisfy the requirements of a producer, in different local production systems, the shelf life and agronomic behavior must also be evaluated. In these cases, the hybrids with better performance had fruit shapes that were close to those of their parent populations from an irregular aspect and produced exceptional commercial-type fruits such as round enlarged (saladette type) or heart-shaped and other commercial types.

In reference to average weight per fruit in this study (52.0 to 192.6 g) was similar to that reported by Mazzucato et al. (2010) in populations of 'A pera Abruzzese' from Italy, ranging from 150 to 366 g, and by Cebolla-Cornejo et al. (2013) with landraces 'Valenciano', 'Muchamiel', 'Penjar' and 'Pimiento', ranging from 113.7 to 302.9 g. In the specific case of 'Muchamiel,' the authors reported that three populations presented yields surpassing 4 kg per plant. The results presented here indicated that is plausible start a participatory breeding program with regional landraces supported by farmers in their own cultivated parcels and principally, with those populations of high yield and fruit quality. Such a proposal is supported by previous experiences such as: Ríos-Osorio et al. (2014), with similar genetic material, landraces from Oaxaca, Mexico, obtained yields up to 8.2 kg per plant in a more intensive production system.

The group of advanced lines presented high weights and uniformity in fruit shape and yields similar to commercial types (2.8 kg per plant). Therefore, these advanced lines are an option for small farmers, which can be used in a combination of alternate parcels or as a varietal rotation with landraces. Thus, farmers could cultivate landraces and advanced lines to diversify crop varieties and as opportunities in regional, national or international markets. In the study region, small and medium farmers are promoting agroecological and organic cultivation; and in these systems, these genotypes are a plausible option and also farmers can produce their own seed.

Hybrids, varieties or advanced lines are commonly evaluated in the practice of the plant breeding of tomato. In this work, we propose a strategy to start participative selection on-farm and in the agroecological conditions of the small-scale farmer when new hybrids or varieties show a decrease in agronomic performance. The advanced lines can compete with new materials because of the approximately 84 g average weight per fruit and more than 30 fruits at the fifth floral branch. Hernández-Leal et al. (2013) found that varieties SUN-7705, Moctezuma and Reserva produced from 99.3 to 117.0 g per fruit and from 1.03 to 1.06 kg of fruit per plant. Riahi et al. (2008) evaluated the varieties and hybrids Rio Grande, Pefectpeel, Hypeel 108 and Firenze and obtained from 56 to 90 g per fruit. Therefore, the agronomic performance of the advanced lines and landraces evaluated in this work is convenient for small tomato producers.

In this work, the first principal component was considered a discrimination index to differentiate genotypes with high performance based on plant growth and physiological traits. A scatterplot of genotypes represented on the two axes is shown in Figure 1; yield per plant and first principal component as the discriminant index. Under these considerations, high fruit yield was associated with taller plants, principally in hybrids H-19, H-06, H-06a and H-69, lines LA-65, LA-113a, LA-113 and LA-108, and two landraces, I-18 and I-35. This result showed that the genotypes of plants with indeterminate growth presented outstanding yields per plant. Later, a second principal component analysis was performed using characters associated to yield (Figure 2), in such case the discrimination among genotypes groups similar to first one, and also it was confirmed that heterogeneous genetic groups were outstanding. Therefore, heterogeneous genotypes (landraces or hybrids) can be an option for small-scale farmers.

Phenotypic homogeneity and uniformity in tomato cultivation are common because of the use of improved varieties or commercial hybrids, which are selected as the goal of a strategy regularly used in plant breeding programs to increase the productivity (Grandillo et al., 1999; Barrios-Masias and Jackson, 2014). With this cultivation approach, the objective is national or international markets for which the quality of fruit is less relevant. However, in recent years, the nutraceutical quality of the tomato fruit has gained major commercial importance, and currently, the quality of fruit is an indispensable character in plant breeding strategies (Grandillo et al., 2011). Moreover, small producers of tomato require varieties or new materials not necessarily with high productivity but with high consumption value associated with flavor, aroma and texture of fruit. Until now, farmers developed or selected new varieties or local varieties from the old varieties, new genetic crossings among commercial varieties and local genotypes or by induced crossing among landraces in which the advanced genotype is highly variable in plant and fruit traits (Mazzucato et al., 2010; Cebolla-Cornejo et al., 2013; Rocchi et al., 2016). Therefore, the results suggest

that it is feasible to select landraces (I-07, I-18 and I-31) or generate non-conventional F_1 hybrids (H-06, H-06a, H-19, H-64, H-67, H-68 and H-69) with high productivity and healthy plants with similar performance to that of advanced lines such as (LA-108 and LA-113a). In the southeast of Mexico, the small-scale tomato producers commonly have a high level of acceptance for highly variable genotypes, and the proposal developed here can be useful for this type of farmer.

All the results in this study were from a greenhouse experiment, as a continuation of previous works developed by Gaspar-Peralta et al. (2012) and Rios-Osorio et al. (2014) using same genotypes at same greenhouse. Therefore, we state that landraces and advanced lines selected in this study as outstanding were also outstanding in previous evaluations, which indicated stability in productivity and fruit size. Consequently, based on the analyses in this study, we can recommend heterogeneous genotypes for selection by small-scale farmers, and when the farmer prefers advanced lines, suggestions can also be provided.

Conclusions

In relevance to producers, breeders, and germplasm curators, we remark that the evaluation of three genetic groups showed significant differences ($p \leq 0.5$) among and within the heterogeneous groups of landraces and non-conventional hybrids and the homogenous group of advanced lines for all evaluated variables, except days after transplant to fruit ripening on the fifth branch. In this study, many outstanding genotypes corresponded to the non-conventional hybrids H-06, H-06a, H-19, H-64, H-67, H-68 and H-69, later landraces I-07, I-18 and I-31 and two advanced lines LA-108 and LA-113a. For the hybrids and landraces, the genotypes had high phenotypic variability in plant and fruit traits.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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