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African Journal of Plant Science

Full Length Research Paper

Allelopathic effect of extracts from selected weeds on germination and seedling growth of cowpea (*Vigna unguiculata* (L.) Walp.) varieties

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***Chromolaena odorata*, *Euphorbia heterophylla* and *Tridax procumbens* are common weeds that are prevalent in cowpea fields. The physiological influence of three dilution concentrations of the aqueous root and shoot extracts of the weeds were examined on seed germination, plumule length, radicle length, fresh and dry weights of plumule and radicle of two varieties of cowpea in the laboratory. The experiment was laid out in a completely randomized design replicated three times. Results revealed susceptibility of two varieties of cowpea (*Vigna unguiculata* L. Walp) (IT99K-573-1-1 and IT07K -292-10) to the allelopathic potential of all the extract concentrations of the selected weeds. Although, all the extracts reduced germination and seedling growth, shoot extracts at 75% concentration of the selected weeds significantly inhibited germination and seedling growth of the variety IT99K-573-1-1 compared with the control which produced 97% (germination %); *C. odorata, E. heterophylla and T. procumbens* shoot extracts produced 22, 20 and 50% germination, respectively. Consequently, *C. odorata, E. heterophylla* and *T. procumbens* shoot extracts produced 25, 18 and 28% germination respectively for variety IT07K -292-10 while the control yielded 99%. Bioassays also indicate that the inhibition was concentration dependent; the inhibition in the extract-treated seeds increased with the increase in the concentration of the extracts. Also, the degree of seed germination inhibition was higher in shoot extracts than root extracts of selected weed. It was clear from the investigation that the extracts of *E. heterophylla* exerted a stronger inhibitory effect on the germination process and seedling growth of the two cowpea varieties than that of *C. odorata* and *T. procumbens*.**

Key words: Allelopathy, allelochemicals, *Chromolaena odorata*, *Euphorbia heterophylla, Tridax procumbens.*

INTRODUCTION

Weed infestation has been known to cause considerable reductions in crop yields thereby hindering sustainable agriculture. Weeds affect crops negatively by competing for nutrients, water, light and space; and also by releasing certain chemicals, which lead to the inter- and intra-plant chemical interactions (Rashed-Mohassel et al.,

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2001). Allelopathy is the ability of a plant to stimulate or inhibit the growth of other nearby plants through the production of allelochemicals (Torres et al., 1996; Javed, 2020). The inhibiting effects of these compounds depend on the concentration received by the affected plants and the susceptibility of the receiving plants (Koocheki et al., 2001). Studies have shown that allelopathy is a major mechanism by which weeds influence the growth of crops (Martin and Smith, 1994). Allelochemicals are most commonly found in plant extracts and in plant residues, some were found in live plant exudates and as volatile gases liberated from leaves (Rice, 1984, Ilori and Otusanya, 2013). Earlier works have shown that allelopathy plays an important role in weed and weed interaction (Kohli et al., 1998; Tajuddin et al., 2002) and weed-crop interaction (Ilori and Otusanya, 2013; Usuah et al., 2013). The study of the effects of allelochemicals produced by certain plants on other plants (weeds on crops, crops on weeds, weeds on weeds and crops on crops) has received increasing attention over the years (Rice, 1984, Marcı´as et al., 2004; Vasilakoglou et al., 2005; Dhima et al., 2006). Several studies have shown that growth cessation by allelopathic compounds covered all life stages from seed until plant maturity; seed germination, seedling growth, leaf area, dry matter, fruit production and biochemical constituents are all affected (Ogbe et al*.,* 1994; Ilori and Otusanya, 2013). Weeds are exerting allelopathic effects on crop seed germination and growth by releasing water-soluble compounds into the soil (Singh et al., 2004). *Chromolaena odorata* in the family Asteraceae has been reported to be a highly invasive weed due to its heavy seed production and aggressive growth rate (Zachariades et al*.,* 2009). Tijani-Eniola and Fawusi (1989) reported on the allelopathic activities of crude methanol extract of *C. odorata* on seed germination and seedling growth of tomato (*Lycopersicum esculentus* L.). Large amounts of allelochemicals such as phenols, tannins, flavonoids in the leaves of *C. odorata* were reported to have an inhibitory effect on the growth of many plants in nurseries and plantations (Eze and Gill, 1992). Moreover, Otusanya et al*.* (2015) and Rafiqul- Hoque et al. (2003) found that different concentrations of *C. odorata* leaf aqueous extract significantly inhibited the germination, root and shoot elongation and development of lateral roots of some plants including cowpea *Vigna unguiculata.* Also*, t*reatment with higher concentrations of inflorescence extract of *C. odorata* had more inhibitory effect on cowpea seed germination (Binumol and Santhoshima, 2018). Similarly*,* Muzzo et al. (2018) reported that *C. odorata* leaf extract inhibited seed germination and seedling growth of some varieties of cowpea and pasture species. Prasada et al*.* (2014) investigated the effects of *Tridax* leachate on the growth of *Vigna mungo* L. and discovered that the increase in leachate concentrate was associated with the increased reduction of seed germination and seedling growth of the test crop. Similarly, several studies revealed that inhibitory effect increases with extract concentration; for example, the leaf extracts of *C. odorata* and *Lantana camara* suppressed the growth and germination of *Vigna radiata* seedlings, and the inhibitory effect was directly proportional to extract concentration (Julio et al., 2019). *E. heterophylla* (Milk weed) and other members of Euphorbiaceae family are well known for the production of a large number of secondary metabolites (Dhole et al., 2011; Saeid et al., 2010). *E. heterophylla* had been reported to be a major weed in soybean cultivation in the United States and Brazil; and it is also a major weed of cowpea (Moore et al., 1990; Winkler et al., 2003). The plant has been found to become resistant to herbicides (Falodun and Agbakwuru, 2003). The aqueous and extracts by ethanol of root, stem and leaf of *E. heterophylla* L. were found to have an effect on seed health (Dhole et al., 2013). Soybean and peanut have suffered yield losses of 30 and 50%, respectively, due to the presence of *E. heterophylla* (Bridges et al., 1992; Willard and Griffin, 1993).

Cowpea (*Vigna unguiculata* (L.) Walp.) is a tropical annual herbaceous legume (family Fabaceae) grown predominantly in Africa and is an important staple crop providing an affordable source of protein (Muranaka et al., 2016). Cowpea is the second most important food grain legume crop in tropical Africa including Nigeria, Niger, Burkina Faso, Uganda and Senegal in nearly all Africa countries south of the Sahara (Onwuem and Sinna, 1990). All parts of the cowpea crop are used, because all are rich in nutrients and fiber. Cowpea is an excellent and inexpensive source of protein, fatty acids, essential amino acids, vitamins and minerals (Fagreia et al., 1990), especially for the poor people in the third world. As a legume crop, the cowpea fixes atmospheric nitrogen through symbiotic interactions with soil rhizobia (Sarr et al., 2015). This implies that cowpea does not need to compete with weed for soil nitrogen, yet it is widely reported that its productivity is greatly constrained by weed activity. Previous studies conducted in Nigeria indicated that weeds and other crops have allelopathic effects on cowpea. Kayode and Ayeni (2009) investigated the allelopathic effects of sorghum and rice husks on cowpea and reported that both types of husks significantly inhibited germination and seedling emergence of cowpea, with sorghum having the greatest effect. Masum et al. (2012) studied the effect of *Parthenium hysterophorus* and *C. odorata* on seed germination and seedling growth of maize (*Zea mays* L.), soybean (*Glycine max* L.) and cotton (*Gossypium hirsutum* L.) under laboratory conditions. Significant inhibition of seed germination by these invasive weeds due to their allelopathic potential was reported with the highest inhibitory effect from aqueous leaf extract of *C odorata* on maize. These included the earlier work of Ogbe et al. (1994) on *C. odorata,* Kayode and Adelawo

(1997) on *E. heterophylla* and Kayode (2004) on *Aspillia africana*.

This study was carried out to (i) determine the allelopathic effects of water extracts of both shoots and roots of *C. odorata*, *E. heterophylla* and *Tridax procumbens* on germination, and growth of plumule and radicle of two varieties of *Vigna unguiculata* L.Walp. (ii) screen the extracts from the weeds for their phytochemical constituents and (iii) compare the allelopathic potential of the extracts of both fresh shoot and root of the three weeds.

MATERIALS AND METHODS

This study was conducted at the Department of Botany, Obafemi Awolowo University, Ile-Ife, Nigeria, at latitude 07°30'N- 07°35'N and longitude 04°30' - 04°04'E. Two varieties of cowpea were obtained from the International Institute of Tropical Agriculture (IITA), Ibadan, Oyo State of Nigeria. Milk weed *(E. heterophylla), T. procumbens and* Siam weed (*C. odorata)* seeds were collected from the demonstration farm of Olashore International School, Iloko-Ijesa, Osun State, Nigeria, at latitude 7°39[1] 15.32'N and longitude 4°48[1] 57.4'E.

Preparation of extracts

The preparation of the aqueous extracts of the weed species was carried out as described by Jafari et al. (2007). Roots and shoots of each weed species were collected separately. For each species, 250 g of the fresh shoots and roots were cut into small segments of about four-cm lengths and finely ground with a mortar and pestle. The ground plant material was soaked in 2 L of water for 12 h. The solution was filtered through cheese cloth to remove debris and the filtrate was further filtered through Whatman No. 1 filter paper. The final extract solution, which served as stock (100%) was diluted appropriately with water to give 75, 50 and 25% concentrations of the aqueous extracts; while distilled water was used as the control. The three dilutions of extract from root and shoot of each of the weeds were considered as treatments used in this experiment. The filtrates were kept in the refrigerator for 7 days to maintain its freshness and to prevent degradation of its allelochemicals, after which another set was prepared. Phytochemical screening for alkaloids, phenols, tannins, flavonoids, saponins, terpenoids, and glycoside was carried out according to the methods of Sofowora (1984).

Experimental layout

A total of 24 shoot and root aqueous extracts was used. Four different concentrations of each of root and shoot aqueous extract of each weed species were obtained. These extracts were applied to the seed of the two varieties of cowpea as explained in the Germination Test. Being a laboratory study, the experiment was considered as a 2 x 2 x 3 x 4 factorial experiment laid out in a completely randomized design with three replicates. Significant means were separated using standard error bars in the line graphs and New Duncan Multiple Range test at 0.05 level of probability.

Germination test

Cowpea seeds were decontaminated by soaking for 10 min in 5% sodium hypochlorite, rinsed for five minutes in running water and finally washed in distilled water. Ten uniform sized seeds of cowpea IT99K-573-1-1 and IT07K -292-10 were randomly selected and placed in each of 120 clean, oven dried Petri dishes (100 x 15mm) which were lined with Whatman No. 1 filter paper. The filter paper served as an absorbent for water or the weed extract that was used as treatment. Twelve Petri dishes were moistened with 5 ml distilled water and served as the control while the other 108 Petri dishes were allotted to different weed extracts. Each was moistened with the 5 ml of the extract. The experiment was arranged using a completely randomized design with three replicates. The entire experiments were kept at room temperature (26°C) for nine days. Germination was determined by counting the number of germinated seeds at 24-h intervals over a 9-day period. Germination Percentage (GP) was calculated with the formula:

$$GP = \frac{\text{Number of Germinated Seeds}}{\text{Number of Planted Seeds}} \times 100$$

Emergence of 1 mm radicle length was used as the criterion for germination. Measurement of the plumule and radicle lengths was done on every other day using a meter rule. Fresh and dry weights of radicle and plumule were measured at the end of the experiment using an electronic weighing balance. For dry weight, seedlings were kept in an oven for 48 h at 60°C. Germination reduction caused by weed extract was computed relative to control.

Statistical methods

Data collected were average of three replicates. All data collected were subjected to analysis of variance to test for significant difference among treatments applied, according to Gomez and Gomez (1984) and significant means were separated using New Duncan's Multiple Range Test (NDMRT). Microsoft Excel was used.

RESULTS AND DISCUSSION

Allelochemicals present in the weed extracts

The phytochemical screening of the water extracts of samples of *Chromolaena odorata, Euphorbia heterophylla and Tridax procumbens* is shown in Tables 1 and 2. The results revealed the presence of terpenoid, glycosides, phenols, alkaloids, and flavonoids in the extract of the three weed species used in this study. In contrast, saponin was not found in any of the extracts (Table 1). *E. heterophylla* had significantly highest concentrations of flavonoids (95 mg/l), phenolic acids (81.158 mg/l) and alkaloids (10.57 mg/l). It however contained the significantly lowest concentration of terpenoids (0.346 mg/l) and glycosides (17.552 mg/l) (Table 2). Moreover, *T. procumbens* (41.807 mg/l) and *C. odorata* (38.925 mg/l) had significantly higher concentrations of glycosides than *E. heterophylla*. The concentrations of flavonoids (81.77 mg/l), phenolics (44.323 mg/l) and alkaloids (9.250 mg/l) in *T. procumbens* were not significantly different from the concentrations of the allelochemicals in *C. odorata*. The

Table 1. Different phytochemicals in the root and shoot of extract of *Chromolaena odorata, Euphorbia heterophylla and Tridax procumbens.*

Sample	Flavonoid	Glycosides	Phenolics	Alkaloids	Terpenoids	Saponins	
C. odorata	+	+	+	+	+	-	
E. heterophylla	+	+	+	+	+	-	
T. procumbens	+	+	+	+	+	-	

+, indicates the presence of the phtochemical compounds; -Indicates the absence of the phytochemical compounds.

Table 2. Total concentration of phytochemicals in the root and shoot of *Chromolaena odorata, Euphorbia heterophylla and Tridax procumbens.*

Weed extract	Total flavonoid (mg/l)	Total glycosides (mg/l)	Total phenolics (mg/l)	Total Alkaloids (mg/l)	Terpnoids (mg/l)
C. odorata	73.03^{b}	38.925^{a}	25.840^{b}	9.708^{b}	0.521^{a}
E. heterophylla	95.00^{a}	17.552^{b}	81.158^{a}	10.570^{a}	0.346^{b}
T. procumbens	81.77^{b}	41.807^{a}	44.323^{b}	9.250^{b}	0.416^{b}

Values within columns followed by same letter are not significantly different using the standard error (SE).

highest concentration of terpenoids was found in the extract of *C. odorata* (0.521 mg/l) and was significantly higher than those of *E. heteropylla* (0.346 mg/l) and *T. procumbens* (0.416 mg/l) (Table 2).

Effect of the different extracts on germination percentage of the crop tested

Figures 1 and 2 shows the effects of both the root and shoot extracts of the selected weeds on the percentage germination of the test crop (cowpea varieties IT99K-573-1-1 and IT07K-292-10). The trend of inhibition of germination by the extracts of the three selected weeds was the same. However, there was a significant difference observed between the control and all the treated plants. The germination % of variety IT99K-573-1-1 was significantly reduced by aqueous root extracts of *C. odorata,* which produced 85, 60 and 39 germination % at 25, 50 and 75% extract concentrations, respectively. Subsequently, shoot extract of same weed produced 80, 44 and 22 germination % at 25, 50 and 75% extract concentrations, respectively. Similarly, for cowpea IT07K-292-10, root extract of *C. odorata* produced 88, 66 and 38 at 25, 50 germination % and 75% extract concentrations, respectively. While the shoot extracts of the same weed resulted in germination % of 86, 45 and 25 when treated with 25, 50 and 75% extract concentrations, respectively. Germination % for both shoot and root extracts treated varieties were significantly different at 50 and 75 % extract concentrations (Figures 1 and 2). Similarly, root extracts of *E. heterophyllum* at 25, 50 and 75% concentrations produced 76, 64 and 34 germination %, respectively, of cowpea variety IT99K-573-1-1. While the shoot extract of the same weed produced germination % of 70, 50 and 20 at 25, 50 and 75% extract concentration, respectively. Also, germination % of variety IT07K-292-10 followed a similar trend; root extract of *E. heterophyllum* produced a germination % of 84, 61 and 36 at 25, 50 and 75% extract concentrations, respectively. While the shoot extracts of the same weed resulted in germination % of 64, 42 and 18 when treated with 25, 50 and 75% extract concentrations, respectively (Figures 1 and 2). Both the root and shoot extracts of *T. procumbens* affected both cowpea varieties the same way as *E. hetrophyllum and C. odorata*. Aqueous root extracts of *T. procumbens* produced higher germination % at the three extract concentrations for IT99K-573-1-1 than its shoot extract. Similarly for cowpea IT07K-292-10, root extract of the same weed resulted in a lower germination % than the shoot extract in all dilution concentrations. For all the selected weed species, there was a significant difference between the germination % of all cowpea seeds treated with shoot compared to those treated with root extracts at 50 and 75% concentrations, except for *T. procumbens* treatment. Percentage germination of all plants treated with 25% dilution concentration of root extracts was not significantly different from that treated with same dilution concentration of shoot extracts. In summary, inhibition of germination by extracts was concentration dependent, and also was plant-parts specific (Figures 1 and 2).

Shoot extracts of all weed species significantly reduced germination percentage of all the treated cowpea plants

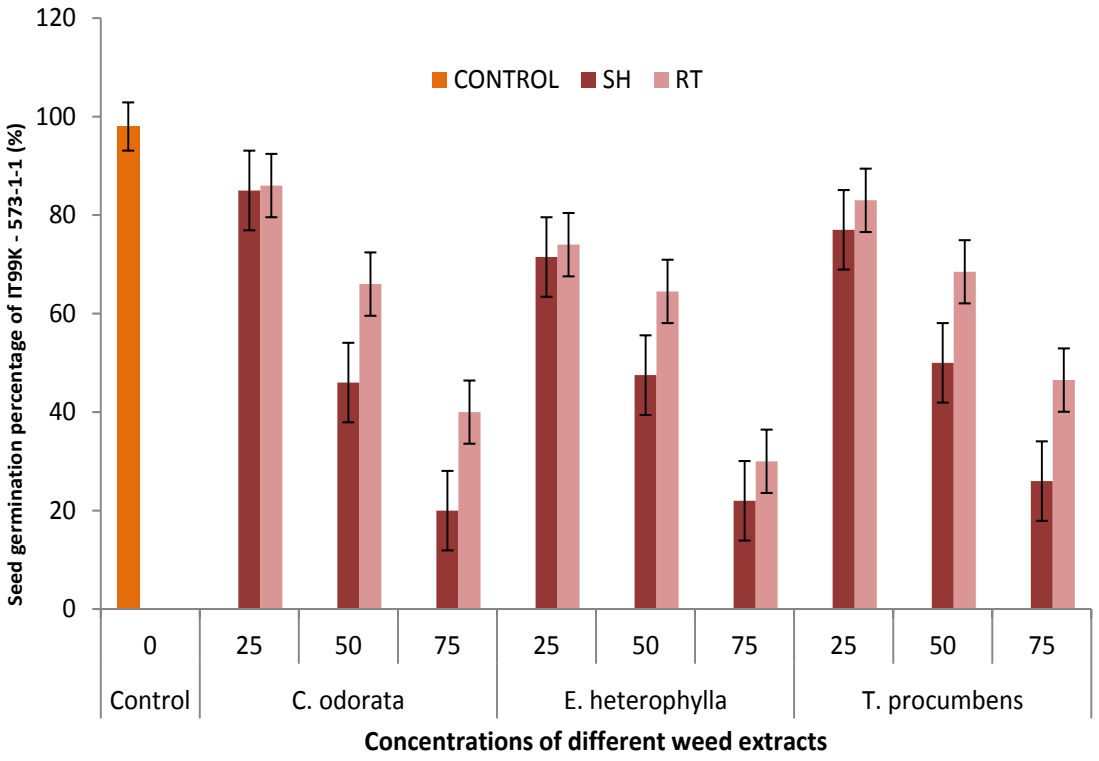

Figure 1. Effects of the extracts of *C.odorata, E. heterophylla* and *T. procumbens* on the percentage germination of the cowpea variety IT99K-573-1-1 under different concentrations of shoot and root extracts. Capped bars indicate standard error bars. SH, Shoot extract; RH,Root extract.

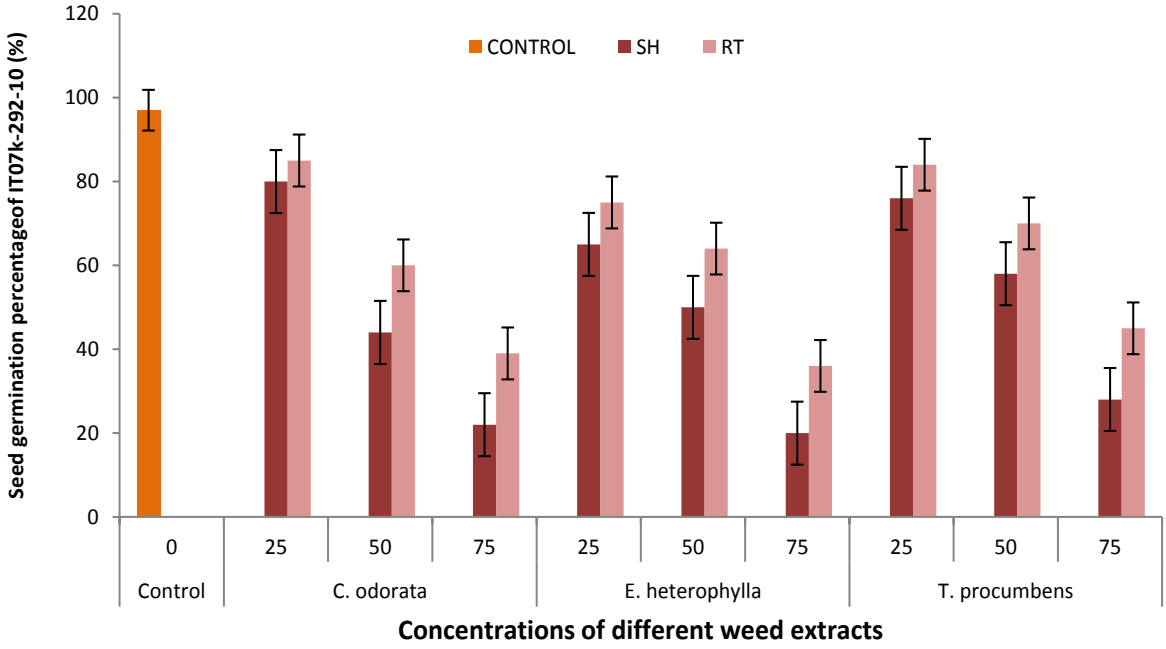

Figure 2. Effects of *C. odorata, E. heterophylla* and *T. procumbens* on the percentage germination of the test crop Variety B: (cowpea variety IT07K-292-10) under different concentrations of shoot and root extracts. Capped bars indicate standard error bars. SH, Shoot extract; RH, Root extract.

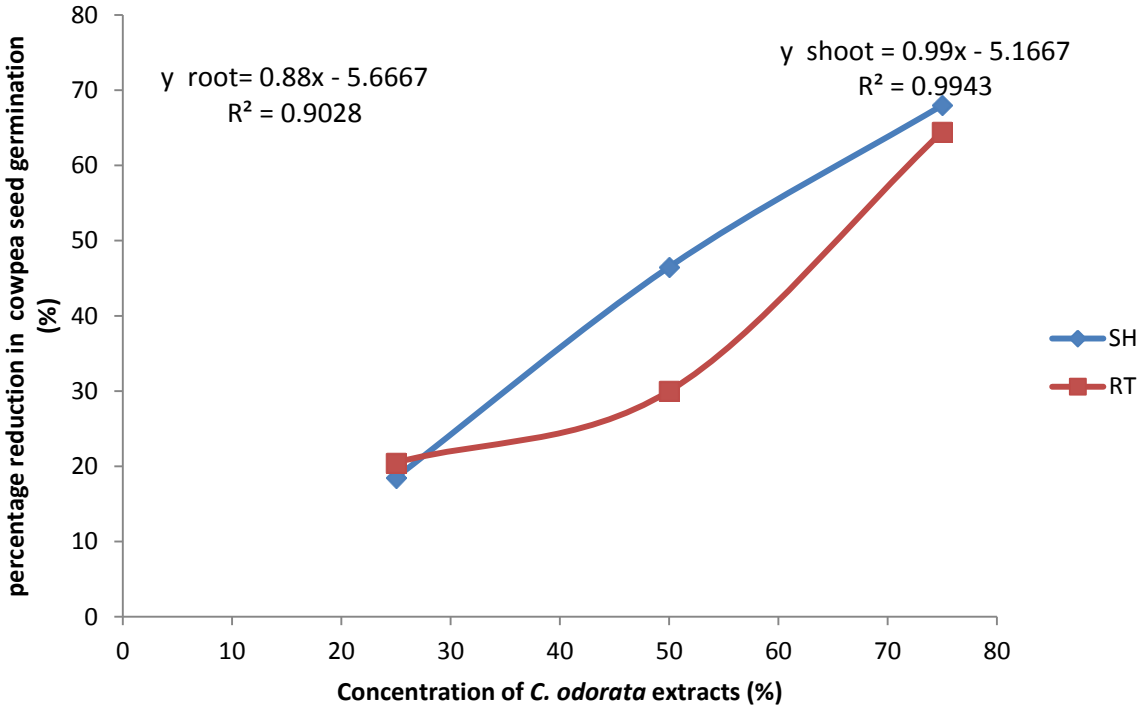

Figure 3. Effect of different extract concentrations of *C. odorata* on percentage reduction in germination of cowpea. Capped bars indicate standard error bars. SH, shoot extract; RH, root extract.

compared to those treated with root extracts. For *C. odorata*, the b-value for the slope of the root extract was 0.88 while that of shoot was 0.99 with very reliable values of coeficient of determination of 90% and 99%, respectively (Figure 3). Similarly, root extract of *E. heterophylla* had a b-value of 0.96 while that of the shoot was 1.2 with coefficient of determination of 99 and 97%, respectively (Figure 4). Also b–value of the root extract of *T. procumbens* was 0.73 and that of shoot extract was 0.76 with coeficient of determination of 99 and 99% respectively (Figure 5). Furthermore, germination inhibition of both cowpea varieties by the selected weed extracts followed the trend: *E. heterophylla* shoot and root extracts significantly inhibited germination followed by extracts of *C. odorata* and *T. procumbens* (Figures 3 to 5).

Effects of the different weed extracts on plumule and radicle lengths

The plumule and radicle lengths of the seedlings of cowpea varieties IT99K-573-1-1 and IT07K-292-10 seedlings in the control were significantly higher than all the other seedlings treated with the extracts of the three selected weeds (Figures 6 and 7) ($P < 0.05$). The control seedling of cowpea varieties had a pooled mean plumule length of 6.6 cm and a mean radicle length of 6.2 cm; while the treated seedlings had a significantly lower plumule length of 3.1 cm and radicle length of 2.9 cm ($P < 0.05$) (Figure 6). The radicle and plumule lengths of cowpea in the control were significantly higher than all the treated plants in both varieties from day five to the end of the experiment; while that of the radicle length was from day six. Seedlings followed the trend of plumule and radicle length increasing with decrease in the concentration of the extracts. Statistical analysis revealed that the extracts had more effect on the radicle length than plumule (Figure 8). Also the degree of inhibition increased with the increase in the concentrations of the extracts.

The process of germination is a crucial stage in plant growth. The extract of the selected weed species significantly retarded the germination of the test crop in this study. The observed inhibition of the seeds of the two cowpea varieties, namely IT99K-573-1-1 and IT07K-292-10 could be attributed to a contribution of allelochemicals present in the extracts of shoot and roots of *C. odorata, E. heterophylla* and *T. procumbens.* Several investigations have shown that the allelochemicals are water-soluble and can accumulate upon release within seeds in direct contact with bioactive concentrations (Winkler et al., 2003; Fara et al*.,* 2014). The result in this study agreed with the findings of Jabeen and Ahmed

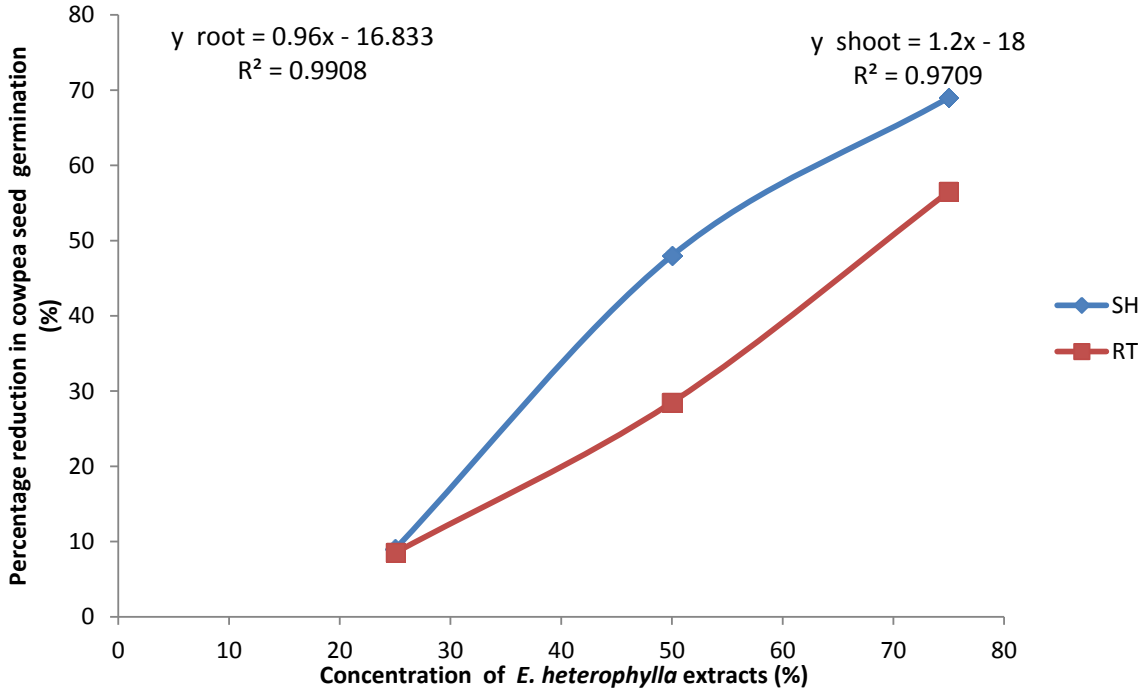

Figure 4. Effect of different extract concentrations of *E. heterophylla* on percentage reduction in germination of cowpea. Capped bars indicate standard error bars. SH, shoot extract; RH, root extract.

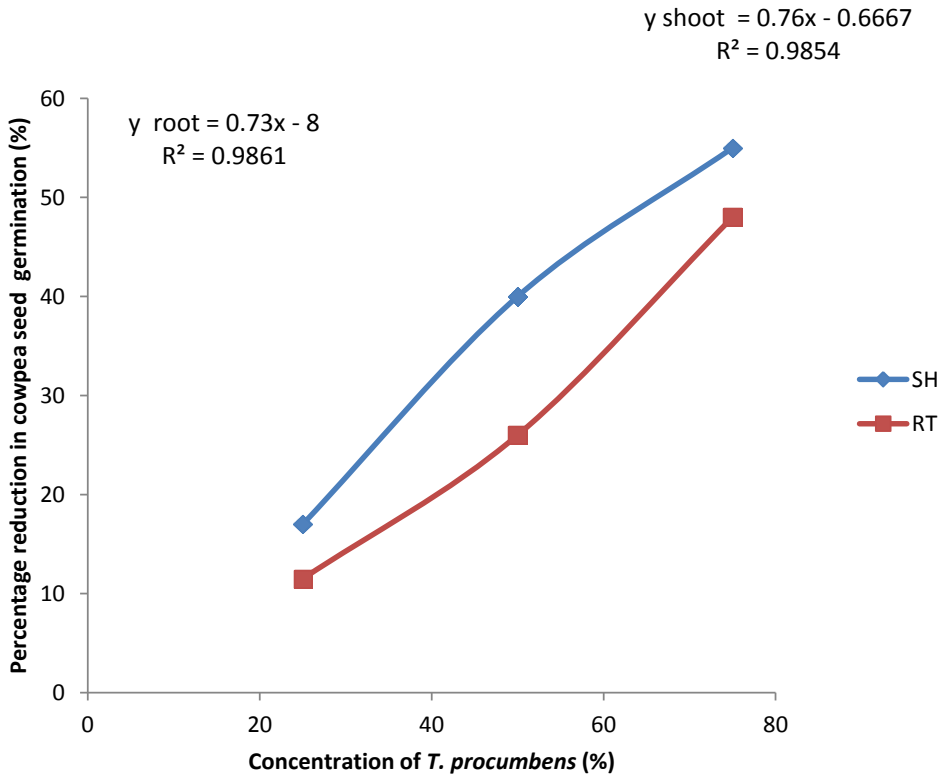

Figure 5. Effect of different extract concentrations of *T. procumbens* on percentage reduction in germination of cowpea. Capped bars indicate standard error bars. SH, shoot extract; RH, root extract.

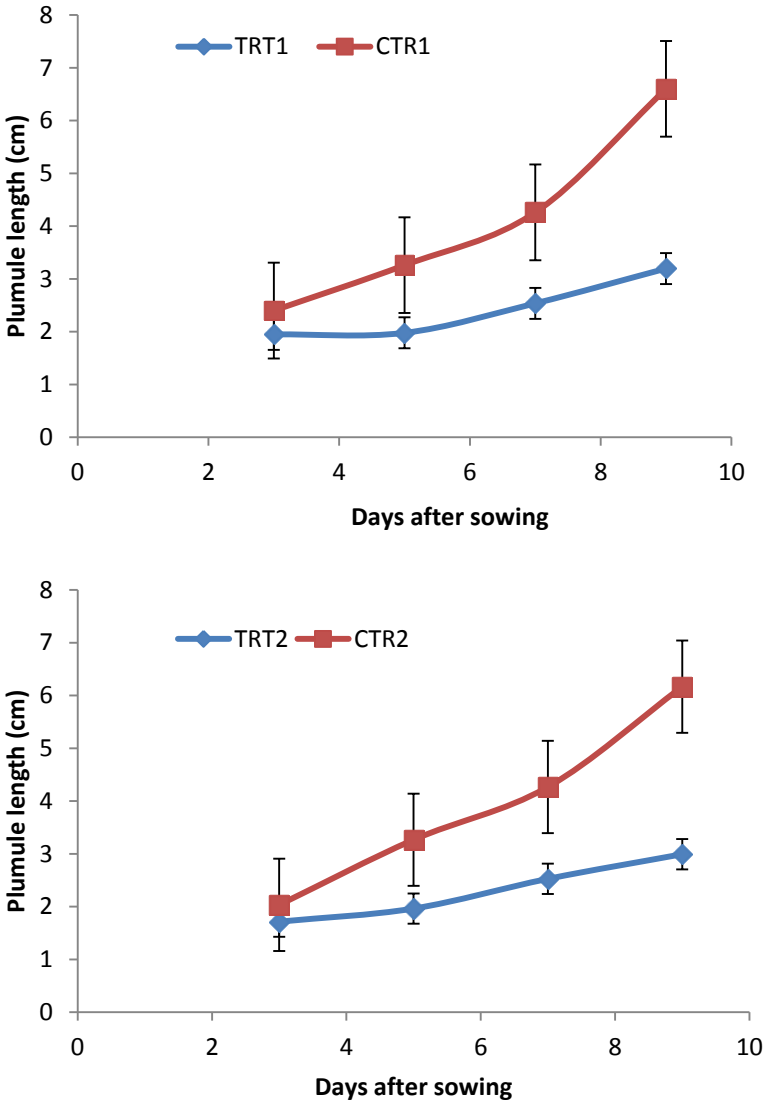

Figure 6. Effect of extracts of the selected weed species on plumule length of the different varieties of cowpea relative to the control (cowpea without extracts application). Capped bars indicate standard error bars. TRT1, Extracts treated variety IT99K-573-1-1; TRT2, Extracts treated variety IT07K-292-10; CTR1, Control for variety IT99K-573-1-1; CTR2, Control for variety IT07K-292-10.

(2009) who observed the effect of *Asphodelus tenuifolius* and *Fumaria indica* on maize seeds and reported inhibition of germination. Moreover, this report is also corroborated by the findings of Oke (1988) that siam weed extract inhibited the germination of seeds of cowpea, soybean and *T. procumbens*. Also, Usuah, et al. (2013) reported that both shoot and root extracts of siam weed inhibited the germination of melon, okra, soybean, cowpea and maize. Dabgar and Kumbhar (2010) found that the aqueous extract of *Euphorbia thiamifolia* inhibited seed germination in *Vigna uriculata* and *Vigna radiata*. The inhibition was also concentration related. The germination percentage of seeds treated with 75% weed extracts was significantly lower than those treated with 50 and 25% extracts, respectively. This finding is similar to the results of Chung and Miller (1995) who found that the degree of inhibition increased with increasing extract concentration. This inhibitory effect on germination of seeds of the species tested in our investigation might be due to allelopathic phytochemicals inhibiting the germination of plants thereby disrupting the cell division, interfering with the mechanism of energy transfer and

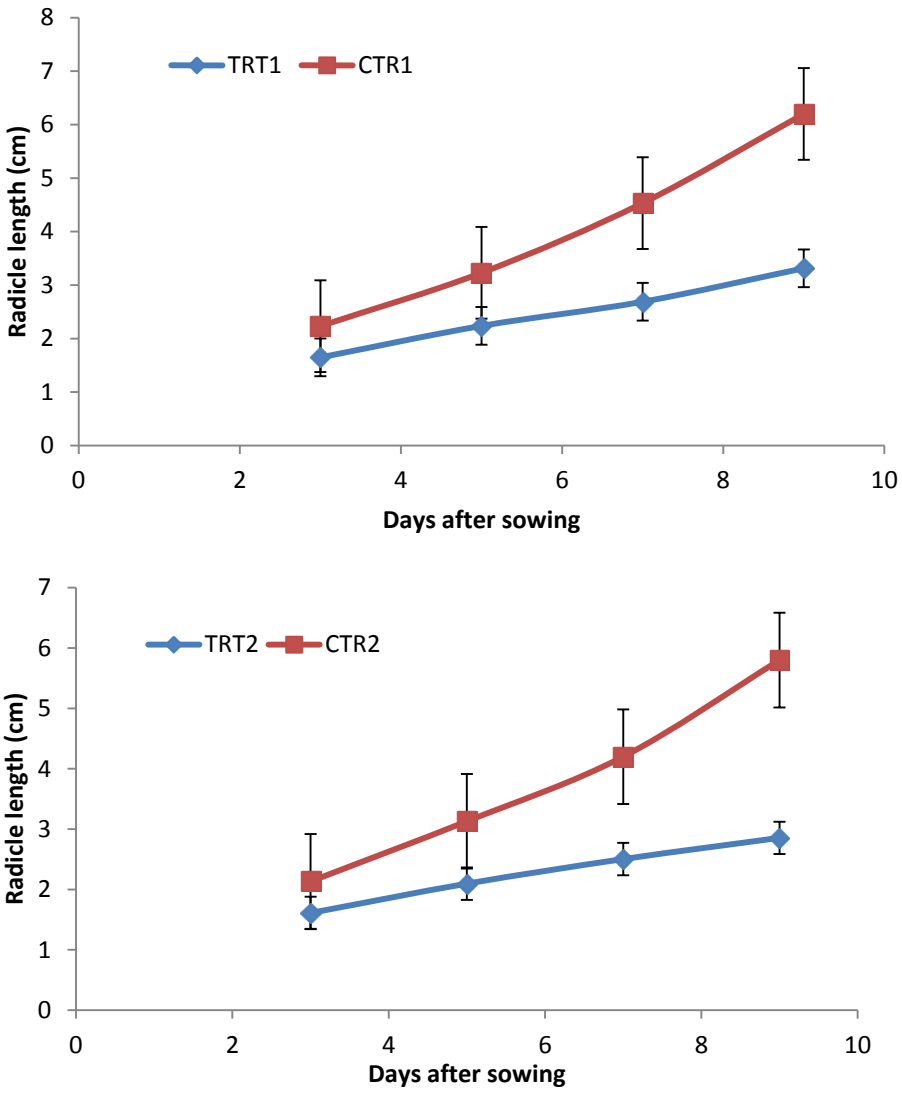

Figure 7. Effect of extracts of the selected weed species on radicle length of the different varieties of cowpea relative to the control (cowpea without extracts application). Capped bars indicate standard error. TRT1, Extracts treated variety IT99K-573-1-1; TRT2, Extracts treated variety IT07K-292-10; CTR1, Control for variety IT99K-573-1-1; CTR2, Control for variety IT07K-292-10.

limiting water and nutrient uptake. Therefore, the inhibitory effect may be due to the entry of water soluble allelochemicals into the seed (Abu-Romman et al., 2010).

In this study, the extracts of the selected weed species significantly inhibited radicle and plumule lengths of the test crop. This is in line with the findings of Kayode and Adanlawo (1997); they revealed that the extracts from leaves of *Gliricidia sepium* had inhibitory effects on the growth of radicle and plumule of cowpea (*Vigna unguiculata*). Allelopathic effect of spurge (*Euphorbia hierosolymitana*) on wheat was studied by Saeid et al. (2010), and they asserted that the extract inhibited both plumule and radicle lengths. Similar result was earlier reported by Kushima et al. (1998) who stated that leachate from melon seeds inhibited the growth of the plumule and the radicle of tomato seedlings. The extracts used in this study had an inhibitory effect on the plumule and radicle length of cowpea varieties IT99K-573-1-1 and IT07K-292-10, which was decreased with increased extract concentration. The results are in agreement with the findings of Jadhar and Goyanar (1992) who noted that the percentage germination, plumule length and radicle length of rice and cowpea decreased with increase in the concentration of *Acacia auriciliformis* leaf

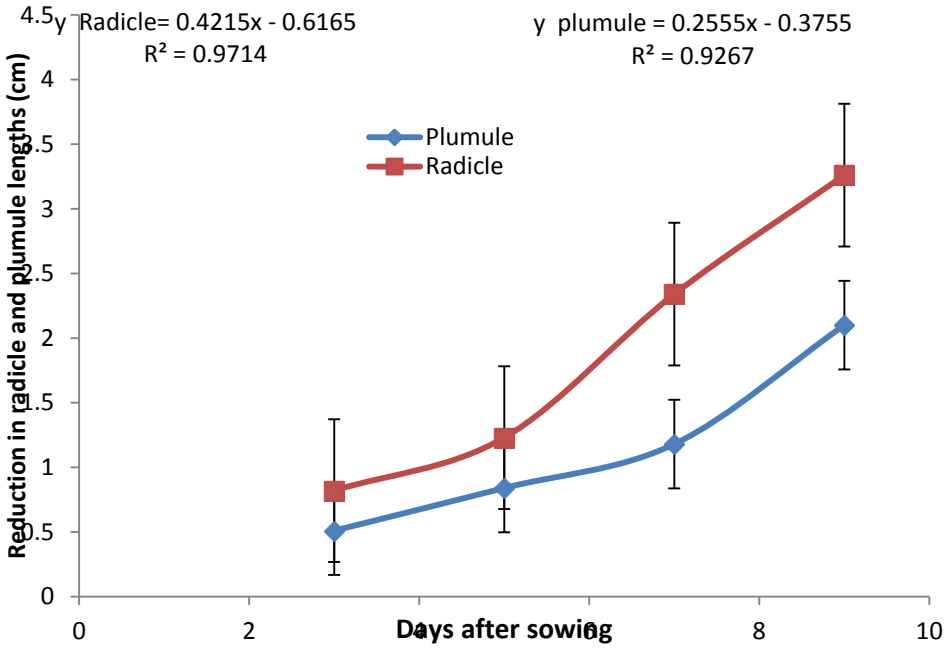

Figure 8. Effect of different extracts of the selected weed species on reduction in plumule and radicle lengths cowpea. Capped bars indicate standard errors.

leachates. Abu-Romman et al. (2010) also reported that the higher concentration of aqueous leachate of *E. hierosolymitana* reduced seed germination, and significantly inhibited the radicle length and growth of wheat seedlings. The reduction in seedling roots length may be attributed to the reduced rate of cell division due to the presence of allelochemicals, which might inhibit gibberellin and indoleacetic acid function (Tomaszewski and Thimann, 1966).

Furthermore, it was revealed that higher concentrations of shoot extracts reduced both the plumule and radicle lengths and was found to be more allelopathic than the root extract. The b-value in this investigation represents the slope of the graph, that is the rate of change in percentage germination due to concentration. The rate of change when shoot extract was used was greater than rate of change when root extract was used. This was collaborated by the findings of Qasem (1995) who reported that shoot extracts were more detrimental than root extracts. In contrast, Rezaie and Yarnia (2009) found both root and shoot extracts to be equally harmful to safflower (*Carthamus tinctorius* L.). The variation in the effect found in this study could be due to difference in phytochemical concentrations in the shoot and root and can be explained by the differences of plant parts in accumulation of phytotoxin (Gonzalez et al., 1997). Also, radicle length was found to be more affected by the extracts than the plumule. This was also consistent with the findings of Friedman (1995), who reported that the allelopathic impact of leachates and extracts are more harmful to the radicle. Alam (1990) also asserted that root growth was more sensitive to the increasing concentration of plant aqueous extracts in comparison to the shoot growth and this could be because the roots were in continuous contact with the extracts.

Conclusion

The water extracts of the selected weed species inhibited seed germination, plumule length, radicle growth, and fresh and dry matter production of cowpea varieties IT99K-573-1-1 and IT07K -292-10. Furthermore, allelopathic effects of weed extracts are weed specific and concentration dependent. Among the different extracts obtained from the weed species, *E. heterophylla* had a more inhibitory effect on cowpea seed germination, plumule and radicle growth compared to *C. odorata* and *T. procumbens.* In the case of weed parts, shoot extracts of the various weed species were more harmful than other root extracts.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Review

A review of plant characterization: First step towards sustainable forage production in challenging environments

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This review paper attempts to give account of how plant characterization assists the availability of information on desirable plant traits, to enhance selective breeding for environmental stresses and thus attain sustainable forage production. Plant characterization is referred to as an account for heritable characters varying from agronomical, morphological to molecular markers. It simplifies grouping of accessions, development of core collections, identification of gaps and retrieval of valuable germplasm for breeding programmes resulting in better insight about the composition of the collection and its genetic diversity. Plant characterization by morphological, physiological and agronomic traits has long been used in selective breeding. Advancement of characterization to the use of molecular markers speed up the process and permits optimal utilization of the adaptive traits harboured in all breeds for stressful environments. In countries like Tanzania, where agro-climatic conditions are challenging, technological progress is slow and market institutions are poorly developed, selecting highly adaptive local varieties is important. Knowledge from characterization of local varieties could be used to breed adaptive and resilient varieties. This will help the farmers to produce enough forage in the fast changing and stressful environmental conditions.

Key words: Characterization, *Cenchrus ciliaris*, drought, salinity, traits.

INTRODUCTION

Livestock producers in developing countries depend on rangelands' forage to feed their stocks (Msalya et al., 2017). The forages from these rangelands are, however, seasonally low in quality and quantity, and therefore negatively influence livestock productivity (Waterman et al., 2007). Butchart et al. (2010) observed that valuable local species are becoming rare, some disappearing or on the brink of extinction. Loss of diversity has consequences beyond just the extinction of species. Once local populations are wiped out, the genetic diversity contained in each species to adjust to environmental stresses is weakened; in turn the livestock production systems are also affected.

Livestock production systems in Tanzania like most

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other developing countries are facing losses in response to a number of drivers (Thornton et al., 2009). Environmental stresses are among the drivers that affect plant and animal productivity (Singh et al., 2011). The effects of these stresses to plants have been documented (Naqvi et al., 2015; Forni et al., 2017; Dzavo et al., 2019). The prevalent ones are those affecting plant water statuses (Claeys et al., 2014). According to Verslues et al. (2006), not having enough water potential for a plant to perform its biological roles can be caused by drought, extreme temperatures and salinity. Plants usually stimulate a complex cellular and molecular mechanism as adaptive response to stresses (Fahad et al., 2015). The United Nation Food and Agricultural Organisation (FAO, 2007) corroborates that wild plants distribution, species' range shifts and gradual biological changes are associated with responses to environmental stresses. Responses of plants to stress differ within and among species depending on stress intensity, stage of plant growth and duration of exposure to stress (Claeys et al., 2014). Availability of varieties and ecotypes of species with different levels of tolerance to environmental stresses is an opportunity for selection and breeding for stress tolerance. Selection and breeding of grasses with stress tolerance traits is inevitable across the globe amid escalating environmental stresses. This paper examines available literatures on plant characterization focusing on sustainable forage production in drought and salinity environments. Several forage species have been characterized but this paper will focus on agro-morphological, physiological and molecular characterization of *C. ciliaris*. The species is selected because it grows well in a wide range of soil types and climatic conditions and it has been adopted by farmers in different regions of Tanzania for pasture establishment. *C. ciliaris* is a perennial deep rooted, tufted and rhizomatous grass, traits which make it fairly adapted to heavy grazing and tolerant to drought (Jackson, 2005; Burson et al., 2012). The grass is wealth of natural ecotypes with morphological diversity which can be visually distinguished and rapidly screened (Burson et al., 2012). Morphological diversity of ecotypes signifies variability of response to environmental stresses hence the need to be characterised.

METHODOLOGY

The information search process was done using Google scholar, an internet- search engines which makes it easy to access online databases. Some of the databases accessed include African Journals Online, Directory of Open Access Journals, Emerald, JSTOR, Research4Life, Science Direct and Web of Science. Furthermore, publications from FAO and International Livestock Research Institute were searched and reviewed. Peer reviewed journal articles, conference papers, government reports, book chapters and thesis published from year 2002 to 2020 were considered for this review. Key words or search terms used were ‘characterization’, ‘plant characterization’, *‘Cenchrus ciliaris’* ‘forage’/ ‘pasture’/ ‘fodder’/, ‘environmental stress’, ‘drought stress’/’water stress’ and ‘salt stress’/ ‘salinity stress’, ‘molecular characterization’, ‘physiological characterization’, ‘agronomical characterization’ and ‘morphological characterization’. Screening of the papers was done by reading the titles followed by the abstracts and where relevant, a full document was read to extract facts, evidences and key messages. A total of 97 publications were recovered of which 19 were reports and book chapters, 78 were journal articles and conference papers. A total of 46 articles were removed of which 10 were abstracts and their full paper could not be accessed, 6 were duplicates and 28 were not relevant, remaining with 51 articles used in this paper.

FINDINGS AND DISCUSSION

The review findings show the description of plant characterization by scholars, approaches of plant characterization, characterization for drought tolerance, characterization for salinity tolerance, plant characterization endeavours in Tanzania, and the novelty of plant characterization.

Plant characterization as concept

There are several definitions or description of plant characterizations given by scholars. However, many scholars refer to it as an account for heritable characters varying from morphological, to molecular markers (Hassen et al., 2006; El-Esawi, 2019). A process which involves recording and compilation of data on important characteristics which distinguish one species from the other and accessions or varieties within species, to enable an easy and quick discrimination among phenotypes (Bioversity International, 2007). Plant characterization reveals desirable traits for both farmers and breeders (Mwenda, 2019; Bucheyeki et al., 2010; Laurentin, 2009). Ability to adapt to environmental stresses, varieties with better quality and high yield are among the desirable plant traits (Laurentin, 2009; Mwenda, 2019).

Approaches of plant characterization

The review reveals that there are several approaches to plant characterization. Substantial works could be sorted in the main approaches, which are agronomic, morphological, biochemical and physiological and molecular characterization.

Agronomic, morphological, biochemical and physiological characterization

Past reviewed works pointed out that visual assessment of growth forms and structure of plants in different types of soils and using variable amount of required nutrients is agronomical and morphological characterization (Jorge et al., 2008; Lima et al., 2018; Wassie et al., 2018). Ago-

Table 1. Phytochemical screening of extracts from different parts of the three ecotypes of *Cenchrus ciliaris* (▲indicates the presence and ◊ indicates the absence of the substances).

Bioactive groups	White			Green			Black		
	Stem	Leaf	Florets	Stem	Leaf	Florets	Stem	Leaf	Florets
Phenol	▲	▲	▲	▲	▲	▲	▲	▲	▲
Flavanoins	▲	▲	▲	▲	▲	▲	▲	▲	▲
Saponins	▲	◊	▲	▲	▲	▲	▲	▲	▲
Glycosides	◊	◊	◊	▲	▲	▲	◊	◊	◊
Steroids	▲	▲	▲	▲	▲	◊	◊	▲	◊
Alkaloids	▲	▲	▲	▲	▲	▲	▲	▲	▲

Source: Kannan and Priya (2020).

morphological traits include plant height, tiller number, tiller type, leaf size and number, internode distance; flower type, size and colour; root type and length. Agro-morphological traits are non-destructive parameters (Fuzy et al., 2019) and they describe plant morphologies efficiently (Blazakis et al., 2017). There are limitations on the use of agro-morphological traits for plant characterization such as limited number of traits to characterize, heritable traits showing insignificant variations and trait expression being influenced by environmental conditions, age and cultivation systems (Blazakis et al., 2017; Laurentin, 2009). Despite the limitations, morphological or visible descriptors remain important for identifying landraces to enhance selection and utilization. These descriptors will continue to be used especially in developing countries until sophisticated methods like molecular markers are easily accessible and affordable. Biochemical characterization refers to characterization based on the types of phytochemicals present in a plant in a given environmental condition, root electrical capacitance, membrane stability index in roots and leave, the amount of methane (CH_4) produced, dry matter and nutrient composition (Kannan and Priya, 2020; Fuzy et al., 2019). On the other hand, characterization of plants based on their functions such as their photosynthesis process, respiration gases produced and nutrient circulation is referred to as physiological characterization (Saini et al., 2007). Furthermore, transpiration rate, CO_2 assimilation rates, is some of physiological descriptors (Fuzy et al., 2019; Mansoor et al., 2015). The limitation of biochemical and physiological descriptors is that they use destructive measurement techniques (Fuzy et al., 2019). Saini et al. (2007) conducted morpho-physiological characterization study with four genotypes of *Cenchrus ciliaris*, two genotypes of *Cenchrus setigerus* and one genotype of *Panicum maximum* grass in an arid ecosystem. In their experiment, seven morphological characteristics and nutritive values were used for characterization. *C. ciliaris* cv. CAZRI 75 had higher total green fodder yield, DM and nutritive value. Based on their results *C. ciliaris* cv. CAZRI 75 was found to be of high potential among the studied grasses to be used in the arid regions of south-west Haryana India. On the other hand, Kannan and Priya (2020) characterised ecotypes of *C. ciliaris* on biochemical compounds found in different components. The ecotypes were grouped based on inflorescence colour variation (white, green and black). According to Kannan and Priya (2020) all ecotypes had the phytochemicals screened regardless of the part containing the compound, with exception of glycosides which were absent in white and black variants (Table 1). Phytochemicals composition is very much affected by environmental stresses, absence or presence of these compounds can be used as a measure the effects of stresses (Daniels et al., 2015).

Molecular characterization

Reviewed studies referred to characterization of organisms using DNA based markers as molecular characterization (Laurentin, 2009, Kumar and Saxena, 2016). The process of molecular characterization used by Ouédraogo et al. (2019) involved DNA extraction and quantification, purifying of the PCR products, sequencing followed by editing of the raw sequences and finally assembly of the readings to check their identity. Molecular markers are prominently used for evolutionary studies, evaluating interrelationship among accessions and among geographical groups. They are also potential for estimation of genetic diversity and identification of duplicates (Laurentin, 2009). It allows simple grouping of accessions, development of core collections, identification of gaps and retrieval of valuable germplasm for breeding programmes, resulting in better insight about the composition of the collection and its genetic diversity (Bioversity International, 2007).

Diversity is important if forage species are to adapt to different environmental conditions and provides a room for selection and breeding. Kumar and Saxena (2016) characterised eight species of genus *Cenchrus* (Table 2) based on their mode of reproduction. There was further characterization using Sequence Characterized Amplified

Table 2. Characterization of Cenchrus species based on mode of reproduction and ploidy status.

Cenchrus species	Accession number	Habit	Ploidy status	Mode of reproduction
C. biflorus	IG-03308	Annual	Diploid (2n = 2x = 34)	Sexual
C. ciliaris	G-693108	Perennial	Tetraploid (2n = 4x = 36)	Apomictic
C. echinatus	IG-96377	Annual	Tetraploid (2n = 4x = 68)	Sexual
C. glaucus	IG-96649	Perennial	Tetraploid (2n = 4x = 36)	Apomictic
C. myosuroides	IG-96380	Perennial	Heptaploid (2n = 7x = 70)	Sexual
C. pennisetiformis	IG-96707	Perennial	Hexaploid (2n = 6x = 54)	Apomictic
C. prieurii	IG-97473	Annual	Diploid (2n = 2x = 34)	Sexual
C. setigerus	IG-01346	Perennial	Diploid (2n = 2x = 36)	Apomictic

Source: Kumar and Saxena (2016).

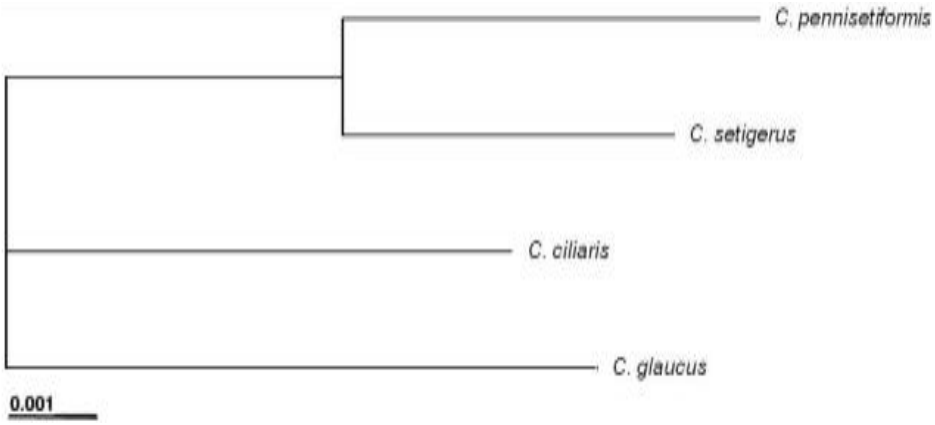

Figure 1. A dendrogram showing sequence diversity among the four apomictic *Cenchrus species* (Kumar and Saxena, 2016).

Regions (SCAR) markers within the apomictic group to establish their diversity. It was observed that *Cenchrus glaucus* had more genetic diversity than the other three species as shown in Figure 1. They concluded based on their research findings that identified markers would be useful for comparative studies and marker assisted breeding of *Cenchrus* (Kumar and Saxena, 2016).

Characterization for drought tolerance

Drought or soil moisture stress is characterised by periods of below average precipitation which are poorly distributed and has become more frequent and enormous problem worldwide (Dzavo et al., 2019; Forni et al., 2017). Drought has negative impact on species diversity, quantity, quality and reliability of forage as well as rangeland vegetation patterns (Giridhar and Samireddypalle, 2015; Nardone et al., 2010). For plants to colonize and continue surviving in drought affected areas need to adapt. Morphological adaptations like development of thick leaves and epidermal layer, waxy cuticle, complex root system and diverse set of molecular mechanisms allow plants to live in extreme conditions (Clauw et al., 2015; Nawazish et al., 2006). According to Acuna et al. (2012) a systematic study of morphological, physiological and biochemical characteristics that provide the ability to tolerate stress can lead to understanding the response of plants to water dearth. Furthermore, Acuna et al. (2012) pointed out traits which can be used in selection for drought tolerance including plant water status, stomata conductance and canopy temperature, spectral vegetation indices, chlorophyll fluorescence and water use efficiency. Understanding desirable traits for high output and drought tolerance is a step towards plant improvement through selective breeding. A study by Mansoor et al. (2002) on 16 biotypes of *C. ciliaris* from the germplasm in Pakistan was conducted to examine the effect of drought on agro-botanical and morpho-genetical characters. Although certain biotypes expressed good individual scores with regard to various characters, one excelled all in plant height, number of leaves, root length and fresh and dry weight. Based on their research findings, Mansoor et al. (2002) concluded that high volume root system is a good index to judge the level of drought tolerance. In another study on root

morpho-anatomical adaptation for drought tolerance in *C. ciliaris,* Mansoor et al. (2015) reported the increase in root number, development of epidermis, endodermis and cortical parenchyma for drought tolerant ecotypes. Drought sensitive ecotypes expressed a decrease in all morphological and anatomical root characteristics (Mansoor et al., 2015). On the other hand, Nawazish et al. (2006) characterised two ecotypes of *C. ciliaris* based on their response to drought by treating them with 100, 75 and 50% field capacity of soil moisture levels. Ecotypes were collected from salt range of Punjab and irrigated soils of Faisalabad in Pakistan. From their results it was shown that the ecotype from salt range adapted better to moderate and high drought levels. The drought adaptive ecotype had increased leaf thickness, cuticle deposition and epidermal layer thickness but had reduced metaxylem area for efficient water transportation in adverse condition. Nawazish et al. (2006) concluded that highly developed bulliform tissue (responsible for leaf culling) and reduced stomata size on the upper surface of the leaf to prevent water loss are important leaf anatomical traits for adaptation to drought. Koech et al. (2014) characterized six range grasses (*Chloris roxburghiana, Eragrostis superba, Enteropogon macrostachyus, Chloris gayana, Soghum sudanense* and *Cenchrus ciliaris*) from the rangelands of Kenya. The aim was to evaluate the effect of different levels of moisture content (80%, 50%, 30% and rain fed) to seed yield of the six species. Among all species characterized, *C. ciliaris* expressed a potential for seed production even under moisture deficit by having no significant difference with the watered treatments (Koech et al., 2014). A good number of ruminants are lost in Sub-Saharan Africa in the periods of drought causing financial loss and food insecurity (Dzavo et al., 2019). Nardone et al. (2010) pointed out that drought affect production in terms of growth, yield and quality of forage produced. The negative effect of drought on forage pose a significant financial burden to livestock producers through decrease in milk component and milk production, meat production, reproductive efficiency and animal health (Naqvi et al., 2015).

Characterization for salinity tolerance

Studies revealed that accumulated salt in soils is a major constraint in agricultural production as it decreases the osmotic potential of the soil with resultant effects on plant water uptake (Roy et al., 2014; Verslues et al., 2006). More than 20% of cultivated land and about 62 million hectare of the world's irrigated soils have been affected by salt (Gupta and Huang, 2014; Fahad et al., 2015). In arid and semi-arid lands, soil salinity is caused by evapotranspiration, lack of leaching water and poor-quality irrigation water (Jouyban, 2012). Resultant effect of soil salinity is visible on seed germination, survival percentage, growth, yield and quality of plants. Report by Verslues et al. (2006) showed that salinity affect metabolism of carbon and nitrogen, assist the accumulation of toxic ions and alter uptake of ions especially K^+ and Ca^{2+} which are important nutrients for plant growth and development. Thus, the effects of salinity on plants can be summarized to include, water deficit due to high pressure on the root zone, ion toxicity and nutrient imbalance (Ronen, 2016). Plants develop physiological and biochemical mechanisms in order to survive in salt affected areas (Gupta and Huang, 2014). Some plant species have moderate salt tolerance and are capable of providing 5 to 10 tonnes of edible dry matter (DM) year^{-1}, at the lower levels of salinity (<15 dS m^{-1}) particularly when the availability of water is high (Masters et al., 2007). Production levels drop and the plant options decrease significantly at high salt concentrations (>25 dS m^{-1}). Al-Dakheel and Hussain (2016) conducted a study in Dubai on genotypic variation for salinity tolerance on 160 accessions of *C. ciliaris.* The levels of salinity used were 10, 15 and 20 dS m^{-1} and 0 for a control and the trait tested for salinity tolerance was biomass yield. Their results revealed that a number of accessions could be grouped in one cluster due to their similar response to salinity levels. Out of 160 accessions characterized, only 12 were stable, salt tolerant and produced a good dry biomass yield, suggesting their potential to contribute to the improvement of grass crops through genetic mechanisms in saline areas (Al-Dakheel and Hussain, 2016). There have been developments in this area with purposes to produce stress tolerant species. A study by Lopez et al. (2011) targeting to obtain a new *C. ciliaris* germplasm that would tolerate salinity and drought was conducted through induced physical and chemical mutation and invitro selection. 500 mature seeds of *C. ciliaris* were subjected to treatment with x-ray (400 Gy) or ethyl methanesulfonate water solution (EMS), 5.5 mM for 24 h. After 7 days germinated seeds were subjected to NaCl and mannitol to simulate salinity and drought respectively. 20 seedlings grown from seeds treated with x-ray tolerated up to 200 mM NaCl, 8 tolerated up to 100 mM mannitol. 21 seedlings grown from seeds treated with EMS tolerated up to 200 mM NaCl and 5 tolerated up to 100 mM mannitol. Hence, a total of 54 tolerant plants were obtained from induced mutation. According to Lopez et al. (2011), 10 plants out of 54 tolerant plants obtained indicated polymorphism with respect to the control cv Biloela using RAPD technique. Only 5 among the polymorphic plants exhibited morphological modification under *ex vitro* conditions.

Studies on forage species characterization in Tanzania

Several studies on plant characterization have been conducted in Tanzania but these studies have been biased on food crops. Gramineae family (a family were

forage grasses belong) selected as examples of characterized food crops in the country are Mangosongo et al. (2019), Dolo (2018), Fisher et al. (2015) and Bucheyeki et al. (2010). Mangosongo et al. (2019) characterized four wild rice populations based on their agro-morphological traits. Their result indicated a wide range of variation for all traits studied among and within populations. The variation in agro-morphological traits presents an opportunity for selection and breeding. A study by Dolo (2018) evaluated the response of 8 rice genotypes to salinity at levels of 0, 50 and 100 mM NaCl. Reduction in physiological traits, ion accumulation and dry matter content of rice were used to distinguish salinity tolerant and susceptible genotypes. The study used marker assisted selection technique to identify salinity associated traits in order to increase selection efficiency and accelerate breeding process. One of the 8 studied genotypes was found tolerant to salinity and was used as a donor parent to improve salinity susceptible genotypes. According Dolo (2018), improved genotypes were more tolerant to salinity than the parent genotypes. Bucheyeki et al. (2010) characterized 40 sorghum landraces from Tanzania and 2 from Zambia. The aim was to determine genetic relationship among landraces and to assess important agronomic traits. Their study observes 78.6% total variability among the races. Bucheyeki et al. (2010) concluded based on their research findings that molecular markers undoubtedly separated landraces within and between groups than morphological markers. On the other hand, a report by Fisher et al. (2015) showed that 25% of maize crop cultivating areas in Africa suffers frequent drought with losses of up to half the harvest. Drought tolerant maize developed after a process of screening, selection and breeding enhanced by information on desirable traits were disseminated to 13 African countries including Tanzania. There was limited adoption of drought tolerant maize seed by famers which varied considerably between countries. Among the factors that hindered fast adoption of drought tolerant maize seed by farmers was limited knowledge on beneficial traits harboured in those new maize seed (Fisher et al., 2015). Farmers and breeders need reliable information to make informed decisions for selection and breeding of forage plants. As it was pointed out earlier, when desirable traits of species, varieties or ecotypes are properly understood, makes a good step towards selection and breeding for environmental stresses. Conversely, there is limited work on range grass characterization for environmental stresses in Tanzania thus deliberate efforts from researchers is required if we are to attain sustainable forage production amid environmental challenges

The novel of plant characterization

With advancement of agricultural and allied science and technology, still the ability to feed the increasing number of both human and livestock in the next twenty years is uncertain, particularly because of the challenging environmental conditions. The demand for good quality soils to produce food crops and fruits has escalated, pushing production of forage crop to marginal lands with a multitude of environmental challenges (Acuna et al., 2012). In this perspective, the ability of forage plants to tolerate environmental stresses is an indispensable character. Various strategies to optimize the reliability and resource use for increased forage demand have been proposed (Busby et al., 2017). Plant characterization is one of priority areas expected to contribute in ensuring adaptive and productive characters are identified and appropriately utilized to enhance plant productivity. Plant breeders will easily access and utilize this information to develop new productive plants of improved tolerance to environmental stresses (El-Esawi, 2019). Govindaraj et al. (2015) suggested that application of plant characterization will lead to long-lasting increased productivity and benefit the environment. It is explicitly that plant characterization can lead to capturing of plant genetic diversity; store it in the form of plant genetic resources like the gene bank and DNA library for long period. The conserved plant genetic resource is readily available materials to be utilized for crop improvement in order to meet future global challenges in relation to food and nutritional security. The use of genetic resources is limited by inadequate essential information on phenotypic and genotypic characters (Shantharaja et al., 2015).

Since plant breeding research and cultivar development are integral components of improving food production, therefore, availability of and access to information on diverse genetic and phenotypic sources will ensure that the global food production network becomes more sustainable (Govindaraj et al., 2015). It was denoted by Hoffman (2010) that, most tropical adapted varieties are essentially uncharacterized and the characterized plants ended at species level documenting a specie's response to different levels and types of stress. On the other hand, there are variations of characters within species important for development of stress resistant cultivars and varieties (Jorge et al., 2008; Acuna et al., 2012). Understanding of adaptation in stressful environments and optimal utilization of the adaptation traits harboured in all breeds needs to be strengthened for the sustainable livestock forages production.

Conclusion

Evidence on substantial decline in livestock feeds quality and quantity due to environmental stresses calls for appropriate strategies to optimize reliability of forage production with scarce resources. Drought and salinity

threaten the sustainability of forage production by negatively impacting plant growth and productivity. A good knowledge of response variation among and within forage species to stress is required to facilitate identification of effective tolerance mechanism. It is worthy taking advantages of available tools and technologies like plant characterization to improve plant selection and breeding targeting adaptable traits to environmental stresses. Agro-morphological, physiological, biochemical and molecular characterization are among the approaches used to generate information on desirable plant traits. *C. ciliaris,* a forage species focused on in this review, revealed its ability to adapt to drought and salinity, stresses which affect water potential for a plant to perform its biological roles. Its adaptation was enhanced by a complex root system, reduced stomata size on the upper side of the leaf, increased leaf thickness, cuticle deposition, epidermal layer thickness and reduced metaxylem area for efficient water transportation. It is through this process that plant characterization fast track systematic information generation to assist plant breeders to efficiently select adapted plants to specific environmental stresses. Limited literature on forage species characterization in Tanzania call for deliberate efforts from researchers in this area.

CONFLICT OF INTERESTS

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Biplot analysis of test environments of quinoa (*Chenopodium quinoa* Willd.) in Burkina Faso

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The identification of stable and adaptable high yielding quinoa (*Chenopodium quinoa* Willd.) and, highly discriminative environments are worthwhile for a successful introduction and adoption of this crop in Burkina Faso. The objectives of this study were to determine the relationship among test environments, to identify the most discriminative and representative test environment(s), and to identify high yielding and stable quinoa variety. The study highlighted that prevailing agrometeorological conditions in an area determine the specificity of the environment. Thus, quinoa growth and productivity is affected by differences in pedological and meteorological conditions. Emerging findings showed that environment E1 at Farako-Bâ characterized by a relative low wind speed (2.03 m/s), no rainfall (0 mm) and moderate temperature (25.07°C), was efficient discriminative and representative of quinoa growing conditions in Burkina Faso for both grain yield and grain yield per plant. Quinoa varieties, Puno and Titicaca were the highest yielding (1132 and 892 kg/ha, respectively) and stable across the environments, while Pasankalla, with an average yield of 779 kg/ha, showed a specific adaptation in two environments having a short day length located at Saria and Lanfiera. The photoperiodicity and temperature were key factors determining the adaptation of this variety in an environment. Plant height and number of branches of Negra Collana were highly stable but its yield performance was low (121 kg/ha). The research implications of this study are numerous, including tailoring quinoa growing calendars and screening a large number of genotypes under the best test environment identified, prior a multi-location trial.

Key words: Quinoa, G x E interaction, GGEbiplot, pedological and meteorological conditions.

INTRODUCTION

Quinoa is an ancestral crop, first domesticated by Andean pre-Columbian tribes 7000 years ago (Babot, 2011). Subsequently, its cultivation spread throughout the region, while diversifying and adapting to new ecotypes:

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Altiplano, Salares, Inter-Andean valleys, Coastal and Yunga zones (Jellen et al., 2015). Quinoa's high genetic diversity is of growing interest in regions where environmental factors can limit the development of crops. Its resilience to abiotic stresses (drought-tolerant, thermo-resistant, halophyte plant, pH versatile, among others) has drawn scientific attention for its potential introduction in marginal environments (Sahel region, Middle East and North African region-MENA) (Bazile et al., 2016a, b). As a result, quinoa´s responses to different growing conditions have been studied worldwide. Changes in abiotic factors like photoperiod, radiations, temperature, soil types, wind and precipitation affect quinoa growth and productivity (Hirich et al., 2012, 2014; Razzaghi et al., 2012; Hinojosa et al., 2019; Alvar-Beltrán et al., 2019; Dao et al., 2020). Variation among quinoa varieties in response to environmental change was also observed. Hence, the investigation of the extent and nature of genotype by environment interaction is crucial for identifying suitable genotypes for a given agro-climatic zone.

Genotype by environment interaction can be explored based on the use of biplots, through Additive Main Effects and Multiplicative Interaction (AMMI) (Gauch et al., 2008) and Genotype and Genotype-Environment (GGE) interaction methods (Yan et al., 2000). The GGE biplot method exploits two sources of variation, GGE interaction simultaneously to evaluate genotype and test environment.

In GGE-biplot different visualization methods were developed to address specific questions relative to genotype by environments data (Yan and Tinker, 2006). Evaluation of both genotype and environment on GGE-biplot allows inspection of various aspects in the adaptability and stability analysis, like discriminating ability and representativeness of environments, mega-environment investigation, 'ideal' genotype identification and which-won-where pattern.

In general, the study of genotype by environment interaction emphasizes the genotype evaluation with identification to high mean performance and stable genotype. However, the environment evaluation is less explored. The understanding of the relationship between test environments can help breeders to reduce the number of environments by selecting one among several environments that provide the same information. Moreover, a prescreening test with multiple cultivars can be carried out in a discriminating and representative test environment for reducing the number of cultivars in multi-location trials. Since its introduction in Burkina Faso, quinoa has been tested in two agro-climatic zones (Soudano-Sahelian and Soudanian zones), for different sowing dates. These growing conditions define different physical environments representative of quinoa cultivation environment in Burkina Faso. Hence, in this study GGE biplot was used to determine the relationship among test environments, to identify the most discriminative and representative test environment, and to identify among four quinoa varieties the one with high yield performance and stable across environments.

MATERIALS AND METHODS

Experimental design

The present study was conducted in Burkina Faso between 2018 and 2019 in five different locations: Farako-Bâ (11°05´N and 4°20´W; 416 m.a.s.l), Saria (12°16´N and 2°09´W; 320 m.a.s.l), Soumousso (11°00´N and 4°02´W; 304 m.a.s.l), Banakeledaga (11°19´N and 4°20´W; 320 m.a.s.l) and Lanfiera (13°00´N and 3°26´W; 358 m.a.s.l). There were 4 sowing dates (from October 2018 until January 2019) at Farako-Bâ and 3 sowing dates (from November 2018 until January 2019) at Saria; whereas the trials at Soumousso, Banakeledaga and Lanfiera were implemented in October 2019. In total, ten different trials were set up. In each trial four quinoa varieties (Negra Collana INIA, Pasankalla, Puno and Titicaca) were randomly laid out in completely block design with three replications.

Management strategies

The soil was prepared manually and prior to sowing the soil was amended using compost at a rate of 5 t/ha (1.1% N content). NPK fertilization (14-23-14) was applied during sowing at a rate of 100 kg/ha, while 30 days after sowing (DAS) urea, $CO(NH_2)_2$ (46% N content), was spread at rate of 100 kg/ha. Prior to sowing the seeds were treated with insecticides (Permethrin 25 g/kg and Thiram 250 g/ kg), and 3 to 5 seeds were introduced per hole at 10 mm depth. At 15 DAS, quinoa plants were thinned and 1 plant was left per hole giving a plant density of 20 plant/m^2. The trials were fully irrigated twice a week using a drip-irrigation system at Farako-Bâ and Saria and furrow irrigation system at Soumousso, Banakélédaga and Lanfiéra.

Agrometeorological measurements

The time to maturity (MAT), plant height (PH), branches per plant (BP), grain yield per plant (GYP) and grain yield per hectare (GY) for the four varieties were collected. In addition, the following meteorological parameters were measured: mean air temperature (°C), precipitation (mm) and photoperiodicity (minutes/day), wind speed (m/s). Prior to sowing, soil samples, were extracted using an auger at 0-40 cm depth to measure the soil physico-chemical characteristics: soil texture (% clay, loam and sand) pH water, organic carbon, organic matter content (%), nitrogen content in the soil (%), while P and K available (mg/kg) for the plant in the soil were estimated using Bray-method.

In this study, 'environment' was defined as the observed microclimate conditions during the period of evaluation. It was characterized by combination of six factors including geographical coordinates, air temperature, photoperiod, wind speed, precipitation and soil characteristics that were prevailing during the trial. Hence, the 10 trials corresponded to 10 different environments.

Data analysis

Analysis of variance was conducted with PROC MIXED procedure in SAS (SAS, 2004) to determine genotype (G), environment (E) and genotype x environment (G x E) effects for all the traits evaluated. Afterwards, data was subjected to genotype, genotype x environment (GGE) biplot analysis (Yan and Tinker, 2006). The

biplots visually describe genotypic performance over multiple environments based on principal components. The biplots were formatted for comparing environments to a hypothetical ideal environment as well as to compare the different genotypes to an ideal genotype. The graphical display of GGE biplot was constructed using GGE-biplot software (GGE-biplot, 2012).

RESULTS

Agrometeorological parameters

The 10 test environments were characterized for having different textural classes: sandy-loam-clay soil for E1 to E7, sandy soil for E8 and E10 and clay-sandy soil for E9 (Table 1). The soil pH ranges for these environments were very strongly acidic (E4; E6), moderately acidic (E1; E2; E3; E5; E7), slightly acidic (E8; E10) and neutral (E9). E8 (0.77%) and E9 (0.69%) showed a slightly higher organic matter when compared to the other environments (from 0.44 to 0.55%). The availability of phosphorus (P) was particularly high in E8 (19.51 mg/kg) and low in E4, E6 (1.18 mg/kg) and E9 (1.91 mg/kg). Whereas, the availability of potassium (K) was higher in E4 and E6 compared to others environments. Analysis of the physico-chemical properties and the texture of the soils of the 10 test environments highlighted the differences among the six soil types, however environments with soil types 1 and 2 presented similar soil composition and texture though they were located distant from each other (Saria and Farako-Bâ). In contrary, environments with soil types 2 and 3 were found in the same location (Farako-Bâ), but showed important differences in soil properties (particularly in terms of pH, P and K availability).

All the environments considered in this study were distributed in two agro-climatic zones, Soudanian agro-climatic zone (all the environments located at Farako-Bâ, Soummousso, Banakélédaga and lanfiéra) and Soudano-Sahelian agro-climatic zone (environments at Saria) (Table 2). Strong Harmattan winds (prevailing winds from the north) were observed in both zones, being more intense in Soudano-Sahelian agro-climatic zone. Slightly variability in wind speeds among the environments in Soudanian agro-climatic zone was observed with E1 recording the lowest wind speed value of 2.0 m/s. There was no a major difference in photoperiod among the 10 environments. The maximum difference in photoperiod between environments was 26 min/day. The shortest day length was in E3 (693.3 min/day) and E9 (690.8 min/day) while the longest day length was in E6 (716.8 min/day) and E7 (715.8 min/day) (Table 2).

Mean-temperatures during the growing period of quinoa were moderate (between 24.4 and 25.5°C in E1, E2, E3 and E9), slightly moderate (between 26.4 and 27.4°C in E4, E5, E8 and E9) and high (above 28.5°C in E6 and E7). Exceptional precipitations were recorded in six environments (E2, E4, E5, E6, E7 and E8), a precipitation of 71 mm registered in E6 was particularly higher compared to others environments. Combined analysis of the geographical coordinates, meteorological parameters (temperature, wind speed, photoperiodicity, and precipitation) and soil properties indicate that each of the 10 environments presented unique characteristics. Only E1 and E2 tended to have similar characteristics.

Mean performance and combined analysis of variance

Environment means grain yield (GY) and grain yield per plant (GYP) ranged from 26.87 kg/ha and 0.23 g/plant, respectively at E6 to 1631.67 kg/ha (GY) at E8 and 16.38 g/plant (GYP) at E3 (Table 3). At E2, quinoa plants were characterized for having a different branching architecture when compared to E5. The quinoa varieties were early maturing (84 days) at E1 and late maturing (120 days) at E6. Genotype means GY and GYP ranged from 0 (Negra Collana, Pasankalla) to 2877 kg/ha and 23.57 g/plant (Pasankalla), respectively. Results of combined ANOVA revealed significant genotype by environment interaction (G X E) effect for all evaluated traits.

GGE plot analysis for maturity day, plant height and branches number

Discriminating ability and representativeness of test environments

The discriminating ability of the environments is indicated by the length of the environment vectors, which is proportional to the standard deviation within the respective environments (Yan et al., 2011). It indicates the ability of the test environment to provide information about genotype difference. Hence, the most discriminative environment has the longest vector.

Discriminating but non-representative test environments are useful for selecting specifically adapted genotypes in a specific environment. The representativeness of test environments referred to the consistency of a targeted environment when compared with the mean of all test environments, it is measured by the angle formed by the test environment with Average environment Axis (AEA), represented by the small circle at the end of the arrow. The smaller the angle between environment vector and the AEA, the more representative the tested environment.

Interpretation of the Figures 1 to 3 indicate that all tested environments were almost equally discriminative for maturity day, plant height and branches number indicating that all these environments detected phenological and morphological difference among quinoa varieties. However, the representativeness of the 10 environments were different. Test environments E1, E2

Table 1. Soil characteristics of the 10 test environments.

Soil type	Environment	Location	*Sowing date	pH (H_2O)	C (%)	Organic matter (%)	N-total (%)	C/N	P_total (mg/kg)	P_available (mg/kg)	K_total (mg/kg)	K_available (mg/kg)	Texture (USDA)
1	E3 –E5- E7	Saria	Nov-Dec-Jan	5.77	0.27	0.47	0.03	9.32	134.49	4.80	1298.21	38.46	Loam-clay-sandy
2	E1 –E2	Farako-Bâ	Oct -Nov	5.78	0.26	0.44	0.03	9.33	126.67	5.35	1460.67	68.89	Loam-clay-sandy
3	E4- E6	Farako-Bâ	Dec -Jan	4.93	0.32	0.55	0.03	11.33	107.00	1.18	2420.33	96.00	Loam-clay-sandy
4	E8	Banakeledaga	October	6.48	0.45	0.77	0.04	11.43	227.25	19.51	745.37	60.95	Sandy
5	E9	Lanfiera	October	6.80	0.40	0.69	0.04	10.46	171.01	1.91	1460.81	56.32	Clay-Sandy
6	E10	Soumousso	October	6.06	0.29	0.49	0.03	10.33	81.74	4.64	1005.53	40.45	Sandy

*Nov: November; Dec: December; Oct: October; Jan: January.

Table 2. Geographical coordinates and meteorological parameters of the 10 test environments.

Environment	Location	Sowing date	Latitude (degrees, minutes)	Longitude (degrees, minutes)	Altitude (m)	Mean temperature (^oC)	Precipitation (mm)	Wind speed (m/s)	Photoperiodicity (minutes/day)
E1	Farako-Bâ	October	11°06’	4°20’	405	25.07	00.0	2.0	698.2
E2	Farako-Bâ	November	11°06’	4°20’	405	24.44	01.5	2.3	696.4
E3	Saria	November	12^o16’	2°09’	300	24.47	00.0	6.5	693.3
E4	Farako-Bâ	December	11°06’	4°20’	405	26.43	30.5	3.1	703.4
E5	Saria	December	12°16’	2°09’	300	26.54	27.4	5.9	701.0
E6	Farako-Bâ	January	11°06’	4°20’	405	28.87	71.0	3.3	716.8
E7	Saria	January	12°16’	2^o09’	300	29.82	43.4	5.6	715.8
E8	Banakeledaga	October	10°20’	4°20’	300	27.35	30.5	2.7	698.8
E9	Lanfiéra	October	13°03’	3°25’	243	25.5	00.0	2.4	690.8
E10	Soumousso	October	11°00’	4°20’	316	27.35	00.0	2.7	696.6

and E3 were the most representative for maturity day, E9 and E10 for plant height and, E7 and E6 for branches number.

The ideal test environments for selecting generally adapted genotype should be both discriminating and representative. In Figures 1 to 3, the ideal test environment is the center of the concentric circles. It is a point on the AEA in the positive direction (most representative) with a distance to the biplot origin equal to the longest vector of all environments (most informative). The closest environment(s) to this point is or are the best. Thus, E1 and E3 are the best environments to determine the maturity period of quinoa varieties, E5, E7 and E10 for plant height and, E6, E7, E8 and E10 for branches number.

Mean performance and stability of the quinoa varieties

Mean performance and stability of the quinoa varieties for maturity day, plant height and branches number is represented in Figures 4 to 6, respectively. The Average Environment Coordinate (AEC) abscissa (single-arrowed line)

Table 3. Mean performances of quinoa varieties evaluated in 10 different environments.

Environment	Variety	Maturity (days)	Plant height (cm)	Branches number	Grain yield per plant (g/plant)	Grain yield (kg/ha)
E1	Negra Collana	103.67	57.24	11.23	2.03	83.46
E1	Pasankalla	91.00	50.67	11.60	2.30	123.71
E1	Puno	70.33	64.17	11.00	2.99	180.83
E1	Titicaca	71.00	50.09	14.17	2.31	180.02
	Mean E1	**84.00**	**55.54**	**12.00**	**2.39**	**147.33**
E2	Negra Collana	112.00	49.57	9.22	1.20	103.52
E2	Pasankalla	98.67	47.68	5.00	2.18	56.71
E2	Puno	73.00	28.98	8.50	4.61	470.18
E2	Titicaca	73.00	51.00	15.00	3.53	273.74
	Mean E2	**89.17**	**45.50**	**9.97**	**2.69**	**212.42**
E3	Negra Collana	110.33	84.55	21.82		82.18
E3	Pasankalla	95.00	95.57	23.53	14.29	2048.28
E3	Puno	70.00	87.42	26.18	16.83	1758.24
E3	Titicaca	67.67	65.80	18.45	18.85	1655.52
	Mean E3	**85.75**	**84.93**	**22.86**	**16.38**	**1386.06**
E4	Negra Collana	138.00	72.33	13.63	1.10	56.42
E4	Pasankalla	123.00	95.91	11.10	0.28	8.34
E4	Puno	76.67	59.70	16.57	4.65	393.85
E4	Titicaca	68.00	66.69	17.40	6.97	613.20
	Mean E4	**101.42**	**71.63**	**15.00**	**3.66**	**315.06**
E5	Negra Collana	138.33	95.47	28.63	0.48	32.07
E5	Pasankalla	121.00	122.63	33.83	1.99	161.60
E5	Puno	76.00	86.93	26.50	1.36	1516.67
E5	Titicaca	68.00	83.40	22.05	1.37	1521.45
	Mean E5	**100.83**	**98.35**	**28.27**	**1.31**	**886.69**
E6	Negra Collana	149.67	80.45	10.70	0.26	25.85
E6	Pasankalla	141.33	98.42	10.95	0.05	3.03
E6	Puno	93.33	81.94	18.50	0.38	61.63
E6	Titicaca	98.00	80.00	17.67		9.03
	Mean E6	**120.58**	**84.00**	**14.77**	**0.23**	**26.87**
E7	Negra Collana	126.67	94.53	12.80	0.00	0.00
E7	Pasankalla	124.33	115.12	7.87	0.00	0.00
E7	Puno	90.00	82.95	28.70	4.55	849.69
E7	Titicaca	95.00	80.00	25.33	0.81	82.68
	Mean E7	**109.00**	**95.49**	**18.68**	**1.39**	**186.47**
E8	Negra Collana	126.00	97.40	9.03	1.75	220.00
E8	Pasankalla	98.00	103.72	8.50	23.57	1543.33
E8	Puno	73.00	86.93	16.60	22.99	2703.33
E8	Titicaca	70.00	75.73	13.83	13.37	2060.00
	Mean E8	**91.75**	**90.95**	**11.99**	**15.42**	**1631.67**
E9	Negra Collana	118.33	94.29	15.21	3.06	346.67
E9	Pasankalla	82.00	87.08	12.63	16.82	2876.67
E9	Puno	74.67	74.81	18.02	17.01	1530.00
E9	Titicaca	73.00	71.43	15.54	9.97	1366.67
	Mean E9	**87.00**	**81.90**	**15.35**	**11.71**	**1530.00**
E10	Negra Collana	138.00	88.17	9.37	8.60	256.67
E10	Pasankalla	99.00	93.50	7.93	12.50	973.33
E10	Puno	78.33	82.05	17.13	8.35	1860.00
E10	Titicaca	75.00	76.77	14.50	6.10	1163.33
	Mean E10	97.58	85.12	12.23	8.89	1063.33

Table 3. Contd.

	Overall mean	96.71	79.46	16.08	6.84	731.2975
	F (G x E)	89.74***	1.95***	7.57***	3.72***	39.26***

*** Significant at the .001 probability.

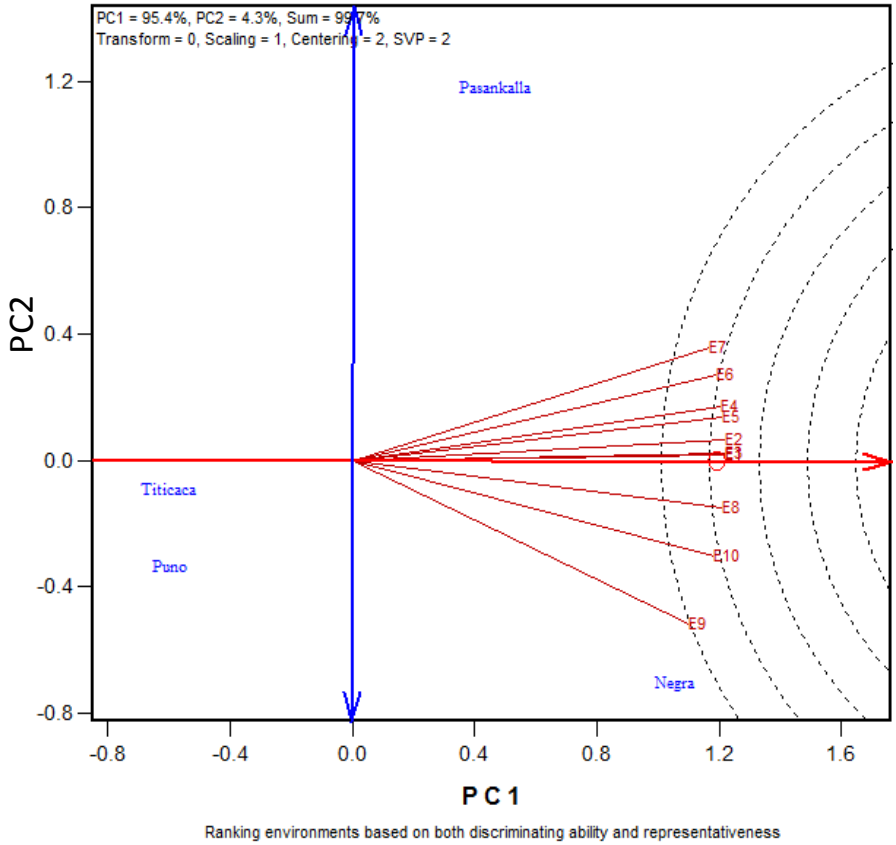

Figure 1. The discrimination and representativeness view of the GGE biplot for **maturity** to rank test environments relative to an ideal test environment (represented by center of the concentric circles).

points to higher mean of the trait across environments whereas the AEC ordinate, double-arrowed line, points to greater variability (poorer stability) in either direction.

Figure 4 represents the time to maturity of the four genotypes in study, Titicaca and Puno were early maturing and consistent across environments when compared to Negra Collana and Pasankalla, characterized for having a longer time to maturity period and for not being stable across environments. The plant height of Negra Collana was very stable across environments contrary to the plant height of others varieties that was very variable (Figure 5). In addition, the graph indicates that plants of Negra Collana and Pasankalla are higher when compared to Puno and Titicaca. Like in Figure 5, Negra Collana had a consistent number of branches across environments (Figure 6) but there was a high variability of branches number for the other varieties especially for Titicaca. Overall, Pasankalla and Negra Collana developed less branches than Titicaca and Puno.

GGE plot analysis for grain yield and grain yield per plant

Correlation, discriminating ability and representativeness of test environments

The correlation between two environments was determined by the cosine of the angle between them.

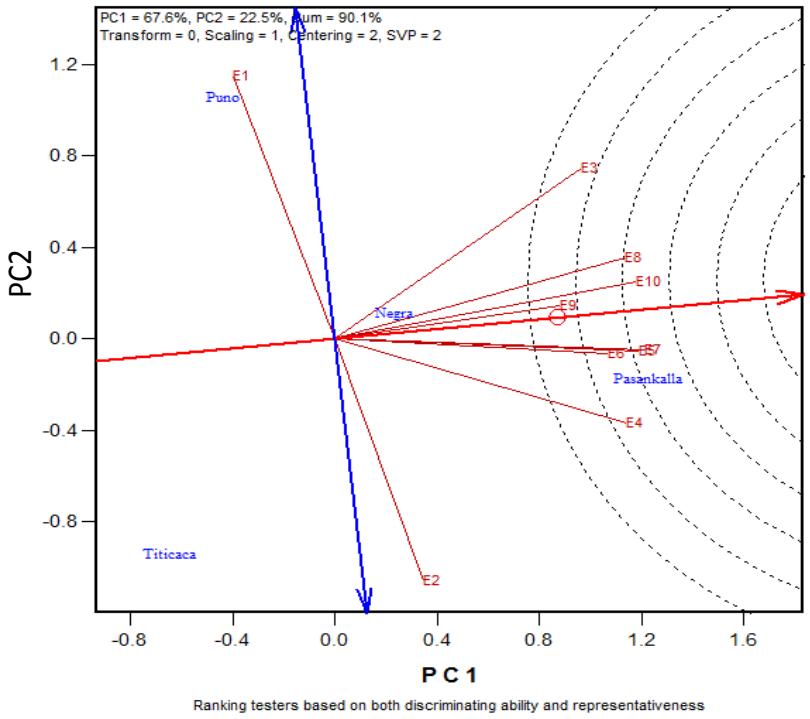

Figure 2. The discrimination and representativeness view of the GGE biplot for plant height to rank test environments relative to an ideal test environment (represented by center of the concentric circles).

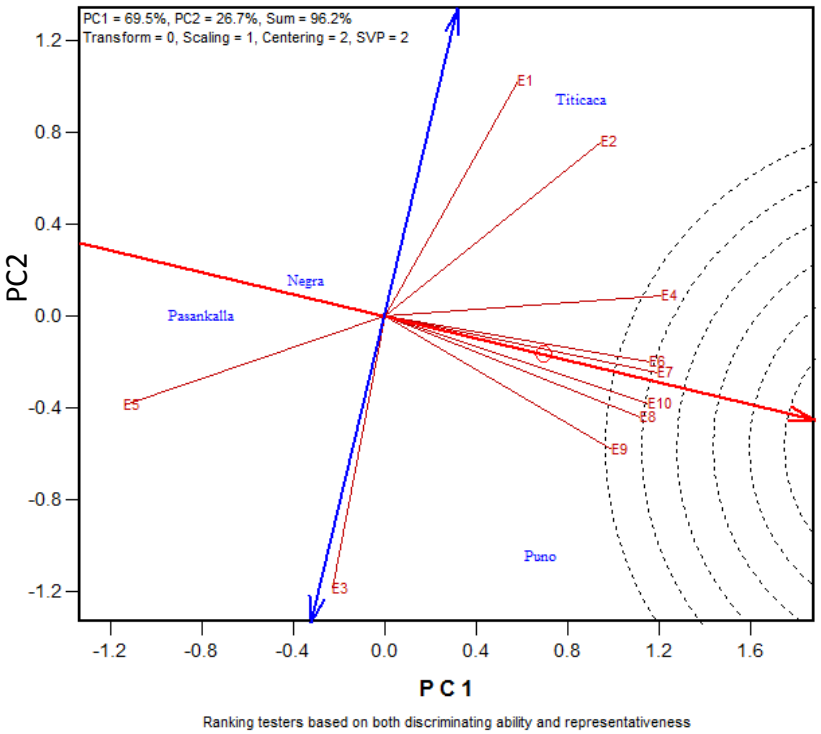

Figure 3. The discrimination and representativeness view of the GGE biplot for branches number to rank test environments relative to an ideal test environment (represented by center of the concentric circles).

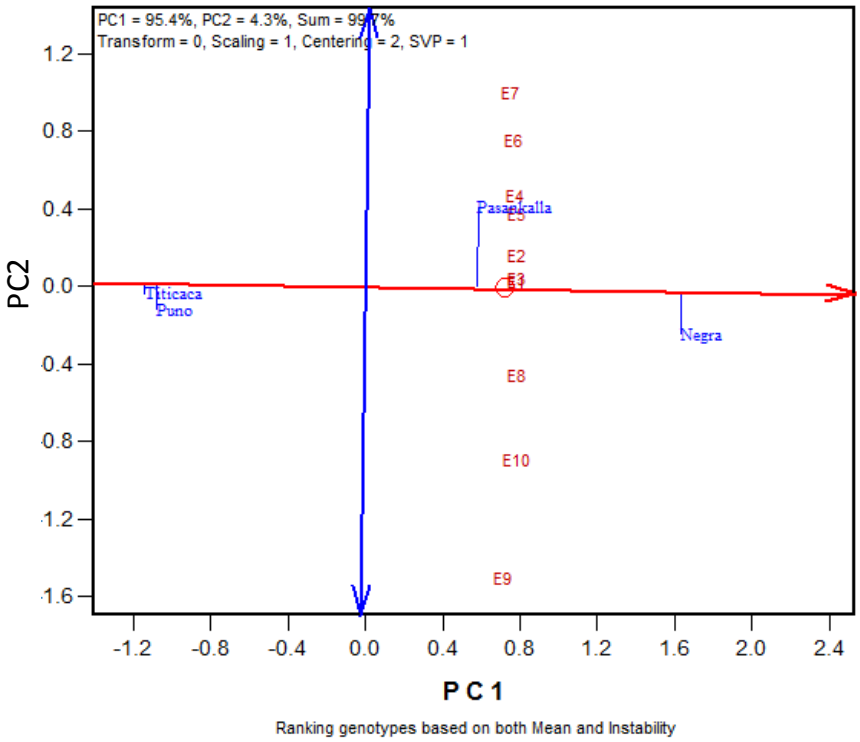

Figure 4. Mean performance and stability of the four quinoa varieties for the maturity day.

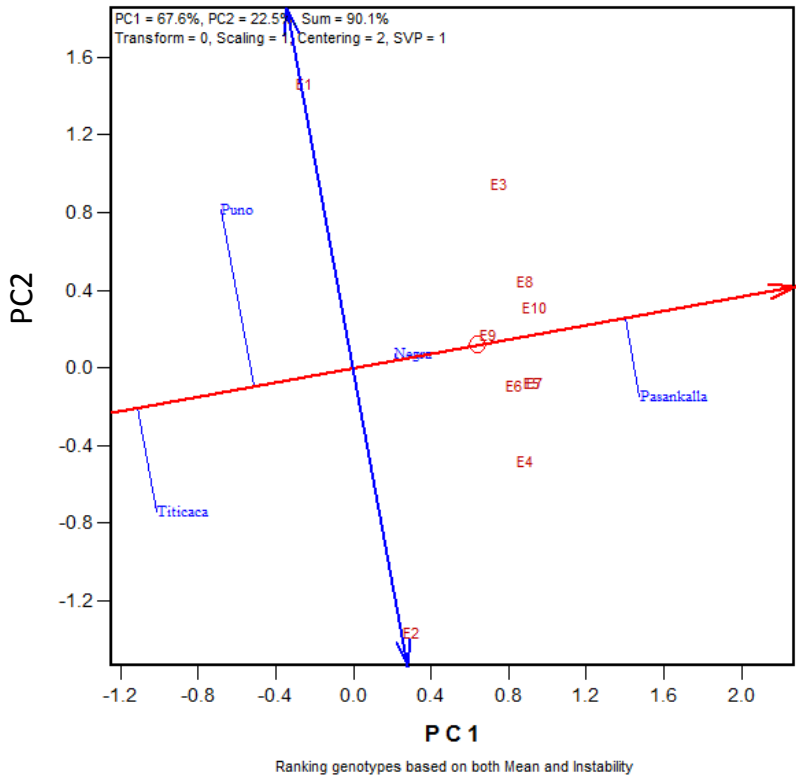

Figure 5. Mean performance and stability of the four quinoa varieties for plant height.

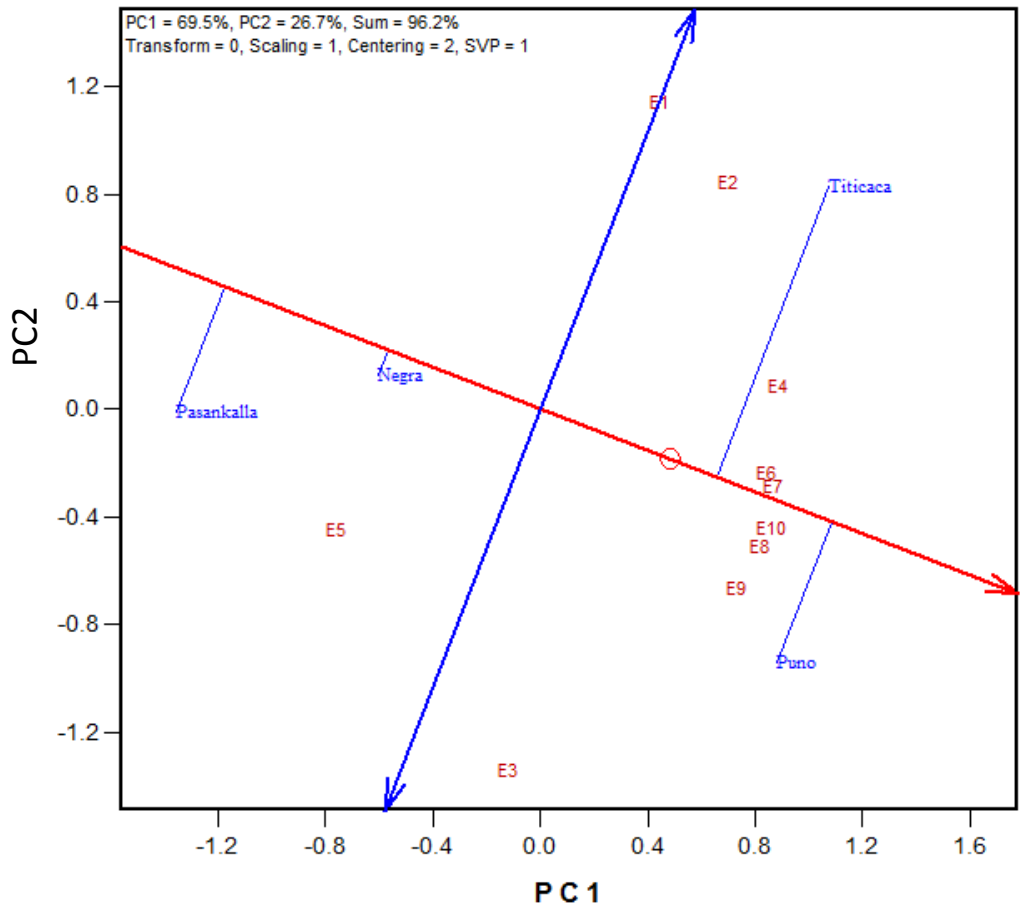

Figure 6. Mean performance and stability of the four quinoa varieties for branches number.

Thus, acute angle (< 90°) indicates a positive correlation, right angle (90°) and obtuse angles (> 90°) denote no correlation and negative correlation, respectively. Figures 7 and 8 indicate three pairs of environments that are closely related. These are E8/E9; E1/E2; E3/E6 for grain yield per plant (GYP) and E1/E10; E4/E5; E2/E7 for grain yield (GY). This indicates that the same information about the yield performance of quinoa varieties could be obtained from fewer test environments, and hence the potential to reduce testing cost.

The graphics indicate also the presence of an important Genotype by environment interaction (GE). This is because of the presence of strong negative correlations (wide obtuse angles) among test environments, which is an indication of strong crossover GE. In Figures 7 and 8 the largest angle are greater than 90° (between E6 and E9 for GY; between E5 and E10 for GYP). The most discriminating (informative) quinoa test environments in Burkina Faso and also the most representative are E1 and E10 for GY and E1 and E2 for GYP. The two environments for each trait are also correlated. E1 is an ideal test environment for both GY and GYP, it should be the test environment of choice to screen quinoa varieties for general adaptability.

Mean performance and stability of the quinoa varieties

GY and GYP of Titicaca were very stable across environments and higher compared to the performance of Pasankalla and Negra Collana (Figures 9 and 10). An “ideal” genotype (the center of the concentric circles) is a point on the AEA (absolutely stable) in the positive direction and has a vector length equal to the longest vectors of the genotypes on the positive side of AEA (highest mean performance). Therefore, genotypes located closer to the ‘ideal genotype’ are more desirable than others.

In this study, Puno was the best genotype carrying both higher yield (GYP and GY) performance and stability and Negra Collana was the poorest genotype. Puno showed a slightly GY variability among environments. Negra Collana and Pasankalla showed a high sensitivity to the environment. GY and GYP variability of Pasankalla were particularly very high.

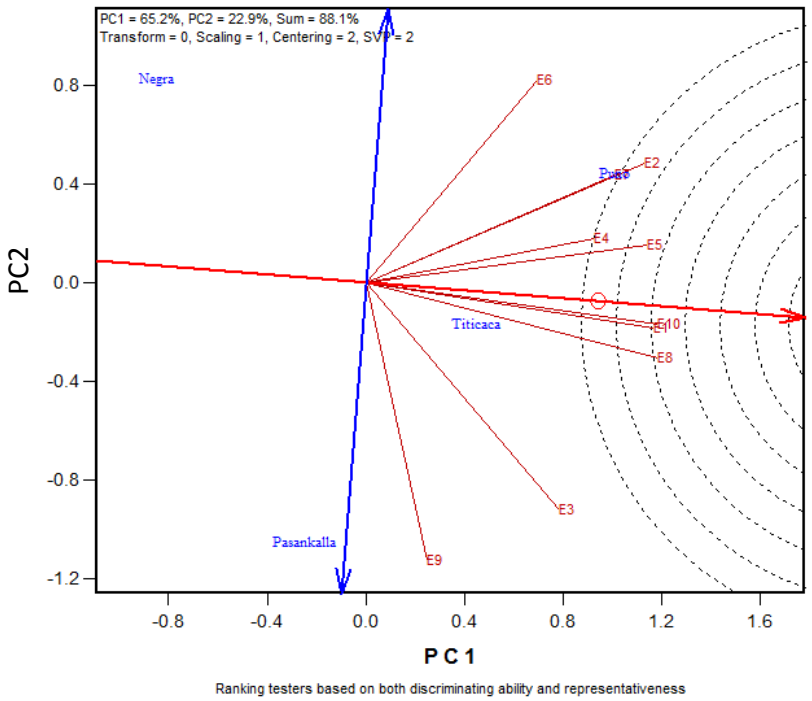

Figure 7. The discrimination and representativeness view of the GGE biplot for grain yield to rank test environments relative to an ideal test environment (represented by center of the concentric circles).

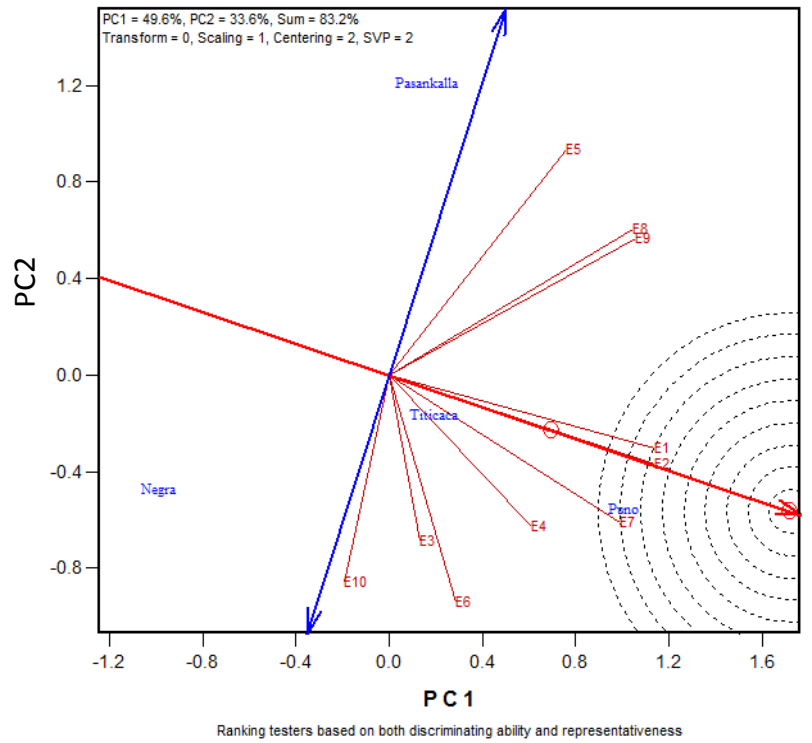

Figure 8. The discrimination and representativeness view of the GGE biplot for grain yield per plant to rank test environments relative to an ideal test environment (represented by center of the concentric circles).

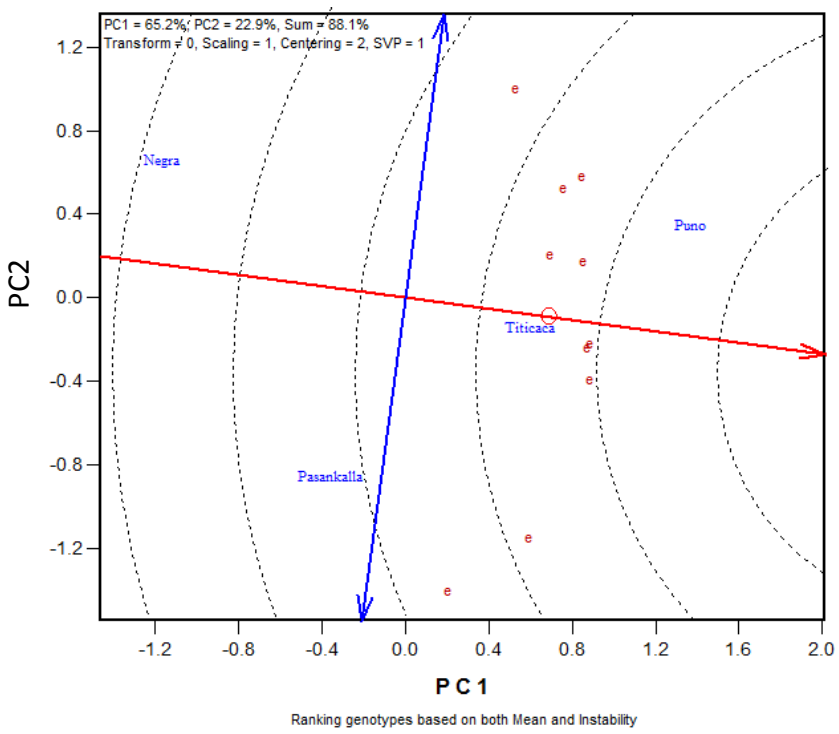

Figure 9. The average-environment coordination (AEC) view to rank the GY of the four quinoa varieties relative to an ideal genotype (the center of the concentric circles).

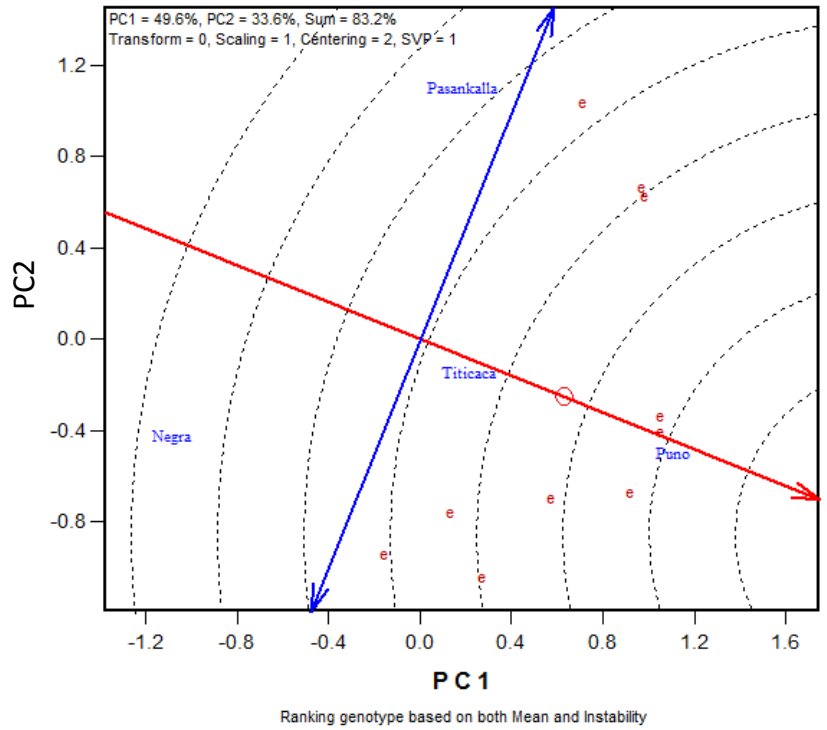

Figure 10. The average-environment coordination (AEC) view to rank the GYP of the four quinoa varieties relative to an ideal genotype (the center of the concentric circles).

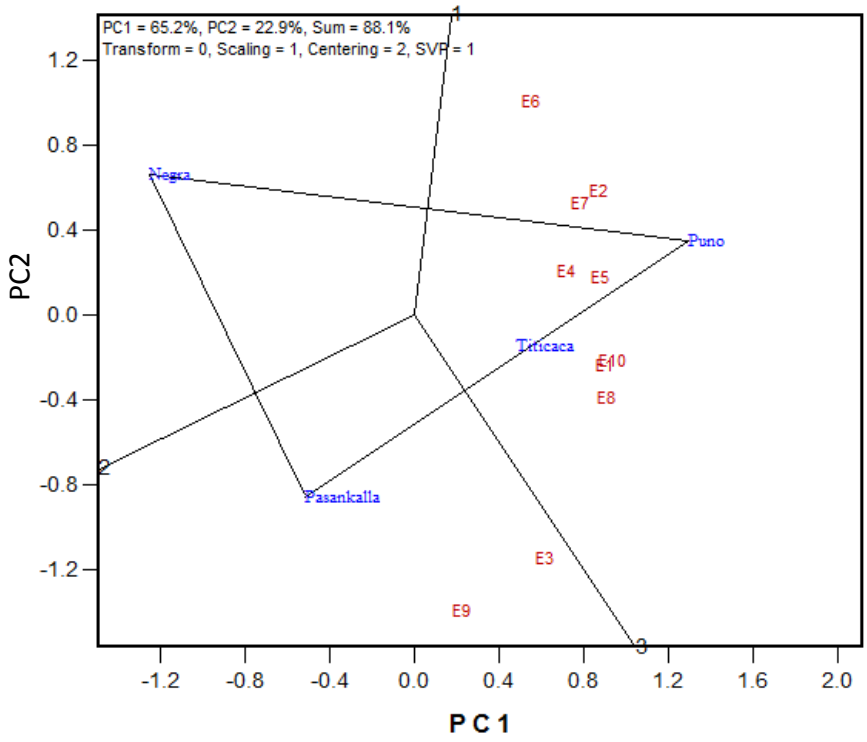

Figure 11. The which-won-where view of the GGE biplot to show which quinoa varieties performed best (grain yield performance) in which environment(s).

Specific adaptation of the varieties (which-won-where)

The function of "which-won-where" of a GGE biplot allows the visual inspection of the mega-environment differentiation and specific adaptation. In Figure 11, there is two representations, a polygon regroup all the four quinoa varieties and three rays that originated from the biplot origin divide the polygon into three sectors. Thus, each sector having its own winning variety at the vertex. Therefore, Pasankalla performed better in environments E3 and E9, whereas Puno was the winning variety in the 8 other environments. Negra Collana was located in a vertex but there was no environment in the sector. It indicates that Negra Collana had the poor yield performance in all the environments. Titicaca is located on the line that connects Puno and Pasankalla showing that the grain yield of the three varieties are ordered as follows Puno, Titicaca and Pasankalla in almost all the environments.

DISCUSSION

Quinoa growth, germination and productivity are affected by several abiotic factors including temperature, photoperiod, soil types, wind and rainfall intensity (Hirich et al., 2012; Razzaghi et al., 2012; Hirich et al., 2014; Hinojosa et al., 2019; Alvar-Beltrán et al., 2019; Dao et al., 2020). The combination of these factors has a major influence on the prevailing agro-climatic conditions of an area.

Results of this study showed correlations among the test environments for all the traits evaluated. However, the environmental factors explaining the similarity of the correlated environments were not clearly established. But, it evidently appears that a combination of several environmental elements determine quinoa growth and productivity. Others abiotic factors that were not collected in this study may also influence quinoa growth. For instance, it was demonstrated that radiation, not considered in this study, influences quinoa growth and yield, with more radiation led to more leaf elongation and growth and consequently decrease in the growing period of quinoa (Bertero, 2001; Hirich et al., 2014). In addition to abiotic factors affecting quinoa growth, the occurrence of some biotic factors such as insect and weeds in the test environments may affect quinoa productivity.

Findings of the study indicated that the geographical position, particularly the Longitude and Altitude, of a location is not a major factor influencing quinoa performance in Burkina Faso. Mukankusi et al. (2016) reported a similar result with altitudinal differences varying from 200 to 900 mm between test environments in a study conducted in Eastern and Southern Africa. However, the Latitude may affect differently the genotypes

depending their sensitivity to photoperiod.

The current study identified the test environment E1 as the best test environment to evaluate quinoa varieties for grain yield and grain yield per plant. It provides more information on the tested genotypes and is representative, to some extent, of the potential quinoa growing environments in Burkina Faso. Quinoa variety selected in such environment will have a large adaptation. The postulate environmental parameters of E1 that made it different to the others test environments are the relative low wind speed (2.03 m/s), no rainfall (0 mm) and moderate temperature (25.07°C). Previous studies demonstrated the influence of these abiotic factors on quinoa growth and productivity. More wind and heavy rain negatively affect quinoa productivity by flattening the plants (Maliro and Guwela, 2015; Dao et al., 2020), and the extreme temperatures are not favorable for quinoa germination, growth and productivity (Garcia et al., 2015; Hinojosa et al., 2018; Dao et al., 2020). These findings suggest that breeders should target such environmental condition to effectively screen new quinoa genotypes in this region.

Results on yield stability of the four quinoa varieties tested suggest that Puno and Titicaca can be recommended in Burkina Faso in all the growing environments since they had both high mean performance and high stability across environments. On the other hand, Pasankalla did not showed a large adaptation but presented a high yield performance in two test environments (E3 and E9), so it should be recommend for specific environments. Pasankalla tends to be highly sensitive to photoperiod than the three others genotypes. The high yields registered for this genotype were 2877 and 2048 kg/ha at E9 and E3, respectively, the photoperiodicity at these two environments was low with 690.8 mm/day and 693.3 mm/day, respectively. However, when the photoperiodicity is high the yield performance of Pasankalla is low. At E7 and E6, it yielded 0 kg/ha and 3.03 kg/ha with a day length of 715.8 mm/day and 716.8 mm/day, respectively. In addition to photoperiod, high temperature and the occurrence of the precipitation at E6 and E7 account for the low yield performance of Pasankalla in these environments.

The visualization of which-won-where patterns of GGE biplot identifies the existence of two different ‘mega-environments’ in quinoa growing conditions. The mega-environment defines by environments E3 and E9, and the mega-environment represented by the 8 others environments (E1; E2; E4; E5; E6; E7; E8; E10). Results showed that all the environments in the latter mega-environment are closely related except E6 evidencing their similarity. On the other hand, the soil and meteorological parameters do not show a clear pattern that could explain the difference of the two mega-environments. However, we hypothesize that the photoperiodicity, temperature and precipitation occurrence might be major factors creating the difference between environments. These findings highlight the fact that mega-environment in quinoa evaluation cannot be define by the physical location or the macro agro-climatic conditions prevailing in a region. For instance, environments E4 and E5 located in Soudanian agro-climatic and Soudano-Sahelian agro-climatic zones, respectively, belong to the same mega-environment. Likewise, environments E3 and E9 forming one mega-environment are located in two different locations and agro-climatic zones.

The morphological and agronomical characteristics of the four varieties have had determined their response to the test environments. Puno and Titacaca are early maturing genotypes with short plants, more branches and a glomerulate panicle while Pasankalla is an intermediate maturing genotype with tall plants, less branches and amarantiform. The poorest variety across the environments was Negra Collana, late maturing genotype with tall plants, less branches and amarantiform. The long maturity period of this variety was the major limiting factor in all the test environments. The genotypes with a compact (glomerulate) inflorescence are more exposed to the effect of winds and heavy rain than a lax (amarantiform) inflorescence. On the other hand, early maturing varieties can easily escape high temperature period contrary to long cycle varieties.

Results on the depiction of the mega-environments and the response of quinoa varieties suggest that crop calendars cannot be tailored according to the different agro-climatic zones in Burkina Faso, opposing to what Dao et al. (2020) recommended. It should rather be adapted to specific soil types and meteorological conditions.

Breeders conduct multi-location trials and employ different G x E analytical methods to identify the most stable genotype for several crops (Mahendra et al., 2016; Mukankusi et al., 2016; Oladosu et al., 2017; Mare et al., 2017; Edmar et al., 2019; Yan and Rajcan, 2002). In addition, these studies help the breeder to select locations that are efficient for distinguishing among genotypes and that are good representatives of the target regions. Identification of redundant test locations can reduce testing cost and improve the efficiency of breeding programs. In the light of this study, multi-location trails can be conducted in fewer locations but in varying the environmental conditions (soil types, meteorological parameters) in a single location. Prior, the major environmental factors affecting the crop in the target region should be identified.

Conclusion

Quinoa, recently introduced in Africa’ Sahelian regions, can contribute to reduce the malnutrition if it is adopted by the population and grown by farmers. Study of genotype by environment interaction pattern provided

useful information on quinoa adaptation in Burkina Faso growing conditions. Soil characteristics, air temperature, wind speed and precipitation were the key agrometeorological parameters identified that characterized quinoa growing environments. The extent of the variation of one or several of these environmental factors will determine the quinoa performance. The variability of quinoa genotypes in response to environmental factors was also proved indicating that the cycle of maturity and plant architecture of quinoa determine its adaptation to an environment. In the light of this study, Puno, Titicaca and Pasankalla can be recommended for release in Burkina Faso. Puno and Titicaca have a large adaption whereas Pasankalla will be recommended for specific environments.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Gene action, combining ability and heterotic performance of Ethiopian Sorghum (*Sorghum bicolor* (L.) Moench) lines under moisture stress areas in Ethiopia

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For sorghum hybrid development, assessment of parental lines is a pre-requisite. However, information on heterotic performance and combing ability of Ethiopian elite sorghum lines is inadequate. ANOVA revealed mean squares had signifying substantial amount of variability amongst genotypes for most traits. Males 72, 81 and 99 were the best performing parents for yield and related traits. Hybrids, 106 x 94, 106 x 90, 106 x 102, 107 x 99 and 107 x 105 were found maximum heterotic hybrids for yield as compare to check. The estimates of variance of combining ability revealed that σ^2gca was found inferior to σ^2sca for all traits except plant height and number of heads. The σ^2gca/σ^2sca ratio revealed preponderance of supremacy gene action for most traits. The degree of dominance was found greater than unity for entire traits except plant height. The estimations of parental GCA effects showed that female 106 and males 79, 96, 94 and 81 were good general combiners for yield and related traits. Based on perse performance, heterotic response, combining ability and nature of gene action for yield and related traits, female parent 106 and male parents 94, 102 and 90 were found most performed. Those parental lines were grouped into dualistic heterotic groups based on their SCA and GCA results.

Key words: Combining ability, elite line, general combining ability (GCA) and specific combining ability (SCA), heterosis, heterotic group, hybrid, sorghum.

INTRODUCTION

Sorghum (*Sorghum bicolor* (L.) Moench) is a diploid C_4 cereal crop which was domesticized in Africa particularly Ethiopia and Sudan. It has 2n = 20 chromosome and genome size of 750 Mb (Paterson et al., 2009). It is grown in highly diverse environments of having water stress, soil fertility and temperature conditions. Sorghum mainly reproduces through selfing with outcross reaching to 15% depending on the nature of head (Pfeiffer et al.,

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2010). In Ethiopia, sorghum grows in the lowland areas which receive lower amount of rainfall and has high temperature whiles the highland is characterized by low temperature and higher amount of rainfall (Mindaye et al., 2016).

Sorghum is the fifth major cereal crop in the world and third in Ethiopia which is the most important dry land crop grown for food, feed, fuel and fodder. Sorghum is generally cultivated for grain and fodder purpose. Besides its traditional uses, it can be used as a raw material for several other alternative products such as starch, silage, syrup, jiggery, alcohol, sugar, wine, vinegar, paper, sweeteners and natural pigments (Bahadure et al., 2016). In Ethiopia, the grain is used for preparation of different local food products that require specific grain quality traits. In some rural areas farmers use the stalk for different purposes; such as fodder to feed their animals, construction materials to build shelter and fences (Mindaye et al., 2016).

Global average grain yield over the years range from 1.42 to 1.45 tones/ha. Meanwhile, in Africa average grain yield of sorghum is 1.62 tones/ha (USDA, 2018). Currently, the average productivity of sorghum in Ethiopia is 2.25 tones/ha, which is higher than the global and continental average grain yield (USDA, 2018). The demand for food grain and feed has shot up significantly with the increase in human population and change of living standards. Hence, there is an increasing demand for technologies that can address these challenges.

For a better breeding program aiming at developing high yielding sorghum hybrids, information about combining ability and heterosis of breeding materials is vital at the early stage. Combining ability and heterosis studies deliver information on the type of gene action governing the inheritance of desirable quantitative traits which enable the breeders to define breeding strategies so as to select suitable parents and hybrids. Sprague and Tatum (1942) identified two types of combining abilities in which they named the average performance of a parent in hybrid crosses as general combining ability (GCA) and the deviation of specific combinations from what is expected on the basis of the average performance of the parents involved in the hybrid formation as specific combining ability (SCA). Therefore, GCA measures additive gene actions while SCA is an indication of genes with dominance or epistatic effects. The estimate of combining ability is useful to predict the relative performance of different lines in hybrid combinations. The information on the nature and magnitude of gene action is important in understanding the genetic potential of a population and deciding the breeding procedure to be adopted in a given population (Ingle et al., 2018).

Potential of sorghum hybrids is estimated from the percentage increase or decrease of their performance over the mid parent (average heterosis) and better parent (heterobeltiosis) (Ringo et al., 2015). Heterobeltiosis is more realistic and practical because it indicates the performance of the hybrid in comparison with the best parent unlike mid-parent heterosis that compares the hybrid with the mean of the two parents. In this study, average heterosis and heterobeltiosis were estimated in order to determine performance of hybrids across dry land environments over their open pollinated inbred line parents. Positive average heterosis and heterobeltiosis in a desired trend is preferred in selection for yield and its components (Sayed, 2016).

Studies have revealed the utility of developing hybrids in sorghum for adoption in the semi-arid tropics of Africa and Ethiopia (Mindaye et al., 2016). These studies consistently identified hybrids that produced more grain yield than the parental lines and local check varieties. However, the hybrids lacked the adaptive traits for diverse local environments; were short and had lower grain extent. The expansion of heterotic pools adapted to a particular environment is one solution to overcome the challenges of end use requirements (Mindaye et al., 2016).

In Ethiopia, sorghum breeding has been mostly restricted to germplasm characterization using phenotypic traits and combining ability tests to assess the heterotic patterns of exotic parental lines (Shimelis and Laing, 2016). Mengistu et al. (2010) evaluate the combining ability of five landraces and one advanced line in half diallel mating design. Similarly, 54 F1 hybrids were evaluated for combining ability effects of major morpho-agronomic traits of introduced parental lines (18 pollinators and 3 A-lines) in 2005 at two drought prone areas (Melkassa and Shewarobit) but none of Ethiopian elite lines were included (Tadesse et al., 2008). So far 108 F1 hybrids were assessed for their combining ability performance derived from six female A-lines and eighteen pollinator lines at three locations (Melkassa, Babillie and Miesso). Among them only three male parental lines were evaluated from Ethiopian developed elite lines while others were introduced from abroad (Egu et al., 2009). In addition to this, a total of 139 F1 hybrids derived from twenty-six lines of eighteen male and eight female lines were evaluated for heterosis and combining ability study in 2013 main season at three testing sites *viz.* Arsinegelie, Bako and Miesso**.** But, only nine local genotypes were used and among them only two local improved inbred lines were used for lowland areas in that study (Mindaye et al., 2016). Even though, high level of genetic diversity was reported (Ejeta, 2007), the potential of new locally developed inbred liens for hybrid cultivar development has not yet been exhaustively assessed. To meet the farmers' demand, it is necessary to maximize the production and productivity by developing varieties or hybrids with high grain and Stover yield through structured, formal and continuous breeding programs. The primary resource of plant breeding programs is the genetic variability available within germplasm closely related to the crop of interest. Currently thousands of inbred lines are found in Ethiopia which is developed in

Ethiopian sorghum research program through a continuous crossing program. These elite lines are not assessed exhaustively for hybrid production since they are progenies of adapted lines that are having good traits including better yield. In general, information on heterotic performance and combing ability on Ethiopian elite sorghum lines is limited.

In this study hybrids and elite lines were evaluated to assess their performance, heterotic pattern and heterobeltiosis for yield and yield components by identifying best heterotic parents and good combiner parents for sorghum hybrid breeding program under moisture stress areas in Ethiopia.

The specific objectives include:

1. To identify promising hybrids under moisture stress environments
2. To determine heterosis and heterotic patterns of locally generated elite lines and establish heterotic groups for future hybrid breeding.
3. To estimate combining ability (GCA and SCA) of Ethiopian sorghum elite lines for important agronomic traits.
4. To estimate heritability of important traits.

MATERIALS AND METHODS

Genetic materials

The experiment was carried out using a total of 70 F1 hybrids which were developed using 2 standard introduced female A-lines (ATX623 and ICSA21) crossed with 35 inbred lines (pollinators) using line x Tester mating design fashion. The male parental lines were selected from inbred lines developed in the Ethiopian sorghum crossing program. These lines were selected based on their adaptive traits to drought stress environment, grain yield performance and above ground biomass production. The hybrids to be developed need to have better biomass to fulfill the interest of farmers. In this experiment, both the test cross hybrids including the male and female parents as well as two hybrids (ESH-1 and ESH-4) and one recently released better biomass producing OPV variety (Argiti) were used as a check. In total the trial comprised of 110 genotypes evaluated in three main dry lowland areas of Ethiopia (Kobo (Longitude 12°08'N, Latitude 39°38'E and Altitude 1479 m.a.s.l) -North Wollo of Amhara Region, Miesso (Longitude 9°14' N, Latitude 40°45'E and Altitude 1394 m.a.s.l) and Erer (Longitude 9°10'N, Latitude 42°15'E and Altitude 1297 m.a.s.l)- West and East Hararghae Zone of Oromia Region respectively) during 2017 main cropping season(summer).

Data collection

The major phenological growth, yield and yield related traits associated with drought tolerance were recorded using sorghum descriptor. These were; days to 50% flowering, plant height (cm), panicle exsertion (cm, NGL (Number of green leaves), chlorophyll content, number of productive tillers, panicle length (cm, panicle width (cm), 1000 seed weight (g), grain yield (kg), stay green score (1-5; 1= for greenness up to 5= for dryness) and other important traits were recorded (IBPGR/ICRISAT, 1993).

Statistical analysis

Analysis of variances

Genotypes were considered as fixed and sites, replications and blocks within replications as random effects. The over location combined analysis was performed using the following linear effects model. The analysis was performed using R: R core team (2018).

$$Yijkl = \mu + gi + sj + (g \times s)ij + r(s)jk + eijkl$$

Where μ is the overall mean, gi is the effect of the ith genotype, sj is the effect of the jth site, (g×s)ij is the interaction effect of the ith genotype by the jth site, r(s)jk is the effect of the kth replication within the j^{th} site and e_{ijkl} is the residual variance.

Estimation of heterosis

Heterosis was calculated as:

(I) Mid-parent heterosis MPH = (F1−MP)/MP] × 100
(II) Better parent heterosis BPH = [(F1−BP)/BP] × 100 and
(III) Economic Heterosis =[(F1-SC)/SC]x100 (Singh and Chaudhary, 1977).

Where F1 is the estimated mean performance of the hybrid, MP, is the average of the estimated performance of the two inbred parents, and BP is the estimated mean values for the better performing inbred parent and SC is the mean value of standard check. The significance was tested using t-test values and the *t* values were computed as: *t* value for mid-parent heterosis (MPH) = $t_{MP} = \frac{F1-MP}{\sqrt{\frac{3MSe}{2r}}}$, *t* value for better parent heterosis (BPH) = $t_{BP} = \frac{F1-BP}{\sqrt{\frac{2MSe}{r}}}$ and *t* value for standard heterosis (SH) = $t_{SH} = \frac{F1-BC}{\sqrt{\frac{2MSe}{r}}}$ (Acquaah, 2012). Then, the calculated t values were compared to the tabular value of t-test at error degrees of freedom corresponding to 5 or 1% level of significance using MS Excel.

Combining ability analysis

Combining ability analysis was performed using the adjusted means for block effects in the individual analyses for traits that showed significant differences among genotypes. The analysis was performed using R: R core team (2018) for individual site and combined across sites. Genotypes were partitioned by mean squares due to hybrids into females, males and females x males effects. The combining ability analysis was used to estimate general combining ability (GCA) effects of the parents and specific combining ability (SCA) effects of the hybrid combinations considering the genotypes as fixed effects. Combined line x tester analysis of variance over locations was carried out for the 70 hybrids and their 37 parents (the standard check was not included). The GCA effect of females and males, the SCA effect of females x males, and their interactions with the environment were determined assuming the model:

$$Y_{ijk} = \mu + g_i + g_j + s_{ij} + l_k + r_{kl} + (g \times l)_{ik} + (g \times l)_{jk} + (s \times l)_{ijk} + e_{ijk}$$

Where, Y_{ijk} = the performance of the hybrid made with i^{th} female and j^{th} males in the k^{th} site, μ = the overall mean, g_i = the effect of the i^{th} females, g_j = the effect of the j^{th} males, s_{ij} = the interaction of the i^{th} females with the j^{th} *males* (effect of the ij^{th} hybrid), l_k = the effect of the k^{th} location, r_{kl} = replication effect

in the k[th] location, $(g \times l)_{ik}$ = the interaction of the g_i and l_k, $(g \times l)_{jk}$ = the interaction of the g_j and s_k, $(s \times l)_{ijk}$ = the interaction of s_{ij} and l_k.

General combining ability (GCA)

General combining ability (GCA) effect of females and males is defined as a deviation of line- and tester-mean from mean of hybrids and calculated using the following equations (Singh and Chaudhary, 1977):

GCA of females = $GCA_i = \frac{X_{i..}}{mr} - \frac{X_{...}}{fmr}$,

GCA of Males = $GCA_j = \frac{X_{.j.}}{fr} - \frac{X_{...}}{fmr}$

Where GCA_i = GCA effect for the i[th] female with ∑ GCAi = 0; GCAj = GCA effect for the j[th] males with ∑ GCAj = 0; Xi. . . = the total of i[th] line over all males (t) and replications (r); X. j . = the total of the j[th] males over all females (fm) and replication (r) and X. . . = the total of all the hybrids over all females (fm), males (m) and replications (r).

Specific combining ability (SCA)

Specific combining ability (SCA) effect of hybrid combinations is the deviation of each hybrid-mean from the mean of all hybrids adjusted for corresponding GCA effects of parents and is computed as:

$$SCA_{ij} = \frac{X_{ij\cdot}}{r} - \frac{X_{i..}}{mr} - \frac{X\ ...}{fmr}$$

Where SCA_{ij} = SCA effect of the ij^{th} hybrid with ∑ S_{ij} = 0 for each j; $X_{i\ j\ .}$ = the total of ij^{th} hybrid combination over all replications (r).

Standard errors for combining ability effects

The significance of GCA or SCA effects was tested by dividing the GCA effects of a line or males and SCA effects of a hybrid by its respective standard error (SE). Therefore, the SE was computed using the following formulae (R core team, 2018):

a. Standard errors for GCA:

For Females $SE(_{gi}) = \sqrt{\frac{MSe}{rm}}$

For Males $SE(_{gj}) = \sqrt{\frac{MSe}{rf}}$

b. Standard error for SCA $SE(S_{ij}) = \sqrt{\frac{MSe}{r}}$

Estimation of variance components for combining ability

The estimates of variance components due to females, males and hybrids were obtained as follows (Singh and Chaudhary, 1977):

$\sigma^2g_i = \sigma^2gca_i = \frac{Mf-Mfm}{r*f}$, $\sigma^2g_j = \sigma^2gca_j = \frac{Mm-Mfm}{r*m}$ and $\sigma^2s_{ij} = \sigma^2sca_{ij} = \frac{Mf-Me}{r}$

Where, $\sigma^2g_i = \sigma^2gca_i$ = Variance due to general combining ability for females $\sigma^2g_j = \sigma^2gca_j$ = Variance due to general combining ability for males $\sigma^2s_{ij} = \sigma^2sca_{ij}$ = variance due to specific combining ability for hybrids; r = Number of replications; f = Number of females; m = Number of males; Mf = Mean square due to females; Mm = Mean square due to males; M fm = Mean square due to hybrids, and M e = Mean square due to error.

Proportional contribution of females, males and their interaction to total variance was computed using the following formula (Singh and Chaudhary, 1977):

$Contribution\ of\ female = \frac{SS_f}{SS_H} * 100$, $Contribution\ of\ Male = \frac{SS_m}{SS_H} * 100$ and $Contribution\ of\ female\ x\ male = \frac{SS_{fxm}}{SS_H} * 100$

Estimation of genetic components

The phenotypic and genotypic variance components and coefficient of phenotypic (PCV %) and genotypic coefficients of variation (GCV %) was estimated using R statistical software version 3.5.1 (R Core Team, 2018). Similarly, broad sense heritability (H^2) for important traits (grain yield, days to flowering, plant height) was computed using the above-mentioned statistical software.

Cluster analysis and heterotic grouping

This analysis was done for grouping of genotypes based on their agronomic performance. This was based on all yield and yield contributing traits. Clustering of genotypes was done using R (R Core Team, 2018) clustering strategy of ggplot2 cluster packages.

RESULTS AND DISCUSSION

Genetic variability and genotype performance across sites

The recorded data on different agronomic traits were subjected to analysis of variance to assure the differences among the experimental genotypes. The combined analysis over locations was done for all traits based on their homogeneity test across locations. The combined analysis of variance for phenological and grain yield and yield-related parameters is presented in Table 1.

The mean square value of combined analysis of variance for grain yield was 2.3 which was highly significantly different (p<0.01). Similarly, the mean square value of inbred parents was 2.4 indicating highly significant difference at p<0.01 between them (Table 1). This shows that the hybrids and the inbred lines have an inherent genetic variability which could be useful to make selection and genetic advancement. The mean squares due to total genotypes for combined analysis were highly significant different at p <0.01 for all the studied traits (Table 1). The analysis of variance revealed significant genotypic effect for all the traits. This provides evidence of the presence of enough genetic variability among females, males, and hybrids and allows further assessment of combining ability analysis.

Table 1. Combined analysis of variance table for performance of all genotypes for yield and yield related traits.

Source of variation	DF	GY	TSW	DTF	PHT	HL	PW	PAS	STG
Sites (S)	2	35.166**	7570.492**	821.461**	48133.220**	213.466**	643.900**	0.152	10.321**
Rep (Site)	3	0.254*	55.155*	280.950**	523.268	99.931**	79.234**	0.373**	3.560**
Genotypes	109	2.489**	31.615**	99.842**	3700.020**	57.092**	6.262**	0.107**	0.196**
Parents	36	2.358**	43.671**	110.642**	3554.017**	39.131**	5.816*	0.082**	0.308**
Females	1	0.101	12.000	147.000	90.750	65.801	0.188	0.000	0.008
Males	34	2.397**	40.243**	96.312**	2749.509	29.657**	5.106	0.070*	0.326**
Females * Males	34	2.046**	24.181	74.574**	430.535	17.994**	4.481**	0.089	0.117
Females vs. Males	1	3.301	191.926**	561.526**	34370.548**	334.605**	35.591**	0.600**	0.015
Hybrid	69	2.341**	26.539**	77.864**	2186.484**	27.017**	6.129**	0.100**	0.147**
Check	2	1.134**	15.292	409.389**	2808.722*	227.476**	0.491	0.394	0.016
Hybrid vs. Check	1	3.300	0.157	0.422	24801.138**	4.503	22.622*	0.102	0.055
Hybrid vs. Parent	1	20.957**	12.124	701.903**	104521.106**	2480.495**	32.441**	0.876**	0.016
Parent vs. Check	1	0.055	0.625	69.429	2043.905	218.420**	7.524	0.398*	0.035
Genotype * S	218	1.854**	19.080	3.889	552.136**	9.425*	4.084**	0.088**	0.118*
Parent * S	72	1.761**	16.827	4.791	513.968**	8.261	4.293	0.064*	0.123
Females * S	2	0.041	1.750	20.250	9.250	11.891	1.313	0.000	0.130
Males * S	68	0.781	17.460*	2.906	519.652**	7.674	4.373	0.067*	0.113
Females * Males *S	68	1.630**	23.773*	2.609	563.082**	9.041	4.171**	0.074	0.113
Females vs. Males * S	2	2.784*	10.371	53.414	825.421	24.592	4.532	0.038	0.454*
Hybrid * S	138	1.905**	20.087	3.140	543.133**	9.855	4.074**	0.092**	0.113
Check * S	4	0.605**	7.833	9.556**	727.306	18.827	1.241	0.159	0.029
Hybridvs Parent*S	2	3.492*	2.558	8.094	2591.617*	6.720	2.762	0.517**	0.457*
Hybrid vs. check * S	2	2.670	71.457*	9.397	264.851	4.275	3.810	0.054	0.085
Parent vs. Check*S	2	2.309	61.378*	4.296	144.334	8.024	4.923	0.151	0.006
Error	327	0.059	15.737	20.210	317.216	7.674	2.838	0.063	0.095

* P< 0.05; ** P< 0.01; CHL = chlorophyll content; DTF = days to flowering; DTM = days to maturity; GY = Grain yield; HE = head exertion; HL = head length; NGL = number of green leaves; NH = number of heads; NPT = number of productive tiller; PHT = Plant height; PW = panicle width; SC = stand count; STG = stay green score; TSW = thousand seed weight.

The genotype x environment interaction showed significant difference for eight traits but there was no significant variation for TSW, DTF and HE (Table 1). The combined analysis also revealed significant differences among parents for all the traits studied. The interaction between test hybrids with environment exhibited significant difference for all measured traits except head exertion. The interaction between parents and the test sites was found to be significant for GY, PHT, DTM, NH, PAS and HE. This suggests that the inbred lines used for the cross development responded differently in different environments. However, the female parents were not significantly different for all the traits measured except for plant height and panicle width. The significant difference in phenological traits (plant height and panicle width) agreed with the established fact that drought hardy crops like sorghum can change their phenological growth in response to the dominant climatic conditions (Tadesse et al., 2008).

Table 2. Group mean comparisons of parents, checks and hybrids.

Statistics	GY	DTF	PHT	CHL	DTM	HL	PW	TSW	NGL	HE	STG
Grand mean	2.5	76.2	175.2	49.1	121.5	26.5	6.7	26.1	7.6	1.1	1.7
Max	4.2	84.0	215.1	55.2	126.4	32.2	10.5	31.9	9.5	1.9	2.2
Min	1.3	66.0	106.0	40.3	104.1	17.6	4.9	19.8	5.0	1.0	1.1
Mean of hybrid	2.7	75.5	185.1	50.1	121.5	27.9	6.9	26.2	7.5	1.1	1.6
Max of hybrid	4.2	81.9	215.1	55.2	125.4	32.0	10.5	31.3	9.5	1.4	2.1
Min of hybrid	1.3	66.2	138.4	45.6	104.1	22.6	5.2	22.4	5.0	1.0	1.1
Mean of male	2.3	78.0	161.6	46.8	121.8	23.5	6.5	26.0	7.8	1.3	1.7
Max of male	3.8	84.0	209.1	51.7	126.4	28.3	8.9	31.9	9.0	1.9	2.2
Min of male	1.3	67.1	123.8	40.3	117.1	17.6	4.9	19.8	6.5	1.0	1.1
Mean of FM	1.8	70.8	108.6	49.6	118.5	28.7	5.0	22.3	8.1	1.1	1.7
Max of FM	1.9	74.5	111.1	50.4	119.9	30.7	5.1	23.3	8.2	1.2	1.7
Min of FM	1.7	67.2	106.0	48.8	117.1	26.7	4.9	21.3	8.0	1.0	1.7
Mean of checks	2.2	82.2	147.6	49.8	122.5	27.7	5.9	26.1	7.8	1.1	1.7
Max of checks	2.6	82.3	165.2	54.2	125.0	32.2	6.1	28.1	8.5	1.3	1.9
Min of checks	1.7	82.1	125.8	44.7	121.1	20.7	5.5	24.9	7.0	1.0	1.6
$LSD_{(0.05)}$	0.6	2.8	17.5	2.4	1.7	2.3	0.9	2.2	0.6	0.2	0.2
SE(m) ±	0.1	0.4	2.3	0.3	0.3	0.3	0.1	0.2	0.1	0.0	0.0
CV%	9.6	4.7	9.8	7.2	2.6	10.0	23.6	14.8	9.9	20.4	20.4

The interactions between the male and female parents were found to be significantly different for eight of the measured traits (GY, DTF, NGL, CHL, DTM, HL, PW and NPT) but showed insignificant difference for the rest of traits. This interaction indicated that male and female parents differed significantly in respect to the main and majority of traits studied in the present investigation. This significant difference of male and female interaction revealed the presence of genetic recombination among the inbred lines which will give rise to better specific combining ability that gives the chance to identify superior hybrids for the desired trait.

The analysis of variance further revealed that hybrids differed significantly for all the traits as their mean square values were highly significant; this point out the existence of considerable genetic variability among the hybrids for all the traits that were studied. Mean squares due to hybrid vs. parent were significant for all traits except TSW and STG. This suggests the presence of heterosis for these traits as parents and hybrids were found to be significantly different (Table 1).

From the combined ANOVA, it was observed that mean squares due to male parents was significant and higher than that of the female parents for all the traits except PHT and PW. This shows the existence of better diversity among the male parents than the female parents for most of the traits measured. The interaction of females with their respected testing sites was insignificant for all the traits. This reveals that the female parents did not respond to the environments differently (Table 1). The interaction of females * males was significantly different for all traits except TSW, PHT, NH, PAS, HE and STG. Similarly, the interaction with the sites was significantly different for GY, TSW, PHT, DTM, PW and NH. Among all the genotypes, checks were significantly different only for GY and DTF.

In general, the significant mean squares of female and male component revealed the presence of additive variance, whereas the non-significant means square revealed the presence of non-additive or dominance variance (Table 1).

Hybrid means performance and magnitude of heterosis across sites

Mean performance of the hybrids

Hybrid as compared to their parents and checks had comparable advantages in mean performance. The mean of grain yield for hybrids ranged from 1.3 to 4.2 t/ha. Among the grand mean of all the genotypes, the maximum GY was attained by a hybrid cross of 106 x 24 (4.2 t/ha) followed by hybrid combination of 106x32 (4.1 t/ha), 106x20 (4.1 t/ha), 107x29 (4.0 t/ha) and 107x35 (4.0 t/ha) with an average value of 2.7 t/ha which had high mean value than the grand mean, mean of checks and mean of parents. Similarly, inbred line parents had mean value of ranging from 1.3 to 3.8 t/ha (Table 2). From the top better genotypes, genotype 24 and genotype 44 were statistically different while genotype 24, 20, 32, 64, 70, 22 and 25 were not statistically different for their GY mean values. But the bottom worst and top better genotypes were statistically different for

Table 3. Mean, range and number of hybrids with positive effect for high, mid parent and better check heterosis (%).

Trait	BPH (%)				MPH (%)				BCH (%)			
	Mean	Max	Min	Hybrids with positive effects	Mean	Max	Min	Hybrids with positive effects	Mean	Max	Min	Hybrids with positive effects
GY	15.7	129	-55	48	32.3	160	-44	59	3	62	-50	36
PHT	15.8	44	-23	62	38.5	59	-3	69	11.8	30	-17	59
TSW	0.7	29	-21	32	9	33	-13	62	11.8	30	-17	15
DTF*	-3.4	19	-16	18	1.4	20	-12	38	-8.2	-1	-20	0
DTM*	-1.7	3	-15	20	1	6	-13	49	-3.9	0	-17	1
PW	7.4	68	-28	44	23.1	89	-12	65	13.3	74	-15	63
HL	2.5	68	-20	34	59.5	236	-10	57	358.2	428	267	70
HE*	-38.6	20	-70	7	-27	24	-59	10	-42	0	-50	10
CHL	1.1	14	-10	42	4.3	16	-9	63	-7.5	2	-16	5
STG*	-7.9	40	-60	25	1.8	68	-55	36	-8	29	-51	24
NGL	-9	6	-39	19	-5.8	11	-34	27	-11.7	12	-41	63
PAS*	-24.3	-6	-46	0	-16.1	5	-40	4	45.6	81	4	70
NPT	12.3	165	-100	23	-1.4	430	-100	29	-79.2	-51	-100	0
SC	-15.7	33	-58	15	-6.7	54	-50	22	-22.2	-4	-57	0

* Those traits which are preferable for the negative effects.

the mean value.

Magnitude of heterosis for hybrids

The phenomenon of heterosis has provided the most important genetic tool in improving yield of crop plants. Identification of specific parental combination capable of producing the highest level of heterotic effects in F1 is of immense value for commercial exploitation of heterosis. Percent heterosis for yield and yield related traits was computed over better parents, mid parent and better check. The magnitude of heterosis varied from trait to trait and even from cross to cross. Heterosis over the mid parent, better parent and better check of the hybrids among 35 inbred lines and two female lines are summarized in Table 3. There was significant variation for levels of heterosis among the parental genotypes. Mean performance of the hybrids and heterosis for grain yield ranged from -43.5 to 160.0 (%) across sites. The best parent heterosis (BPH) for grain yield ranged from -55.2 to 129.4(%) with the mean value of 32.3%. The magnitude of heterosis of the hybrids in relation to the better check hybrid (BCH) ranged from -50.0 to 61.5% with the mean value of 3.0% in combination of all testing sites. In general, 84.3% of the crosses exhibited positive MPH and the rest negative heterosis whereas 25.7% of crosses shows >50% of MPH (Table 3). The BPH analysis for grain yield also was done based on the higher performed parent of Hybrids. 68.6% of hybrids attain positive heterosis for BPH and there also 31.4% of hybrids show negative heterosis from high parent of among the corresponding male and female parents. In general, 10 hybrids get greater than 51% of BPH. The magnitude of heterosis varied from cross to cross and trait to trait. For a specific trait considerable high heterotic effects were observed in certain crosses and low in others, which revealed that nature of gene action varied with the genetic makeup of parents. The results indicated that both positive and negative heterosis was observed for these studied traits. For days to heterosis was desirable but for rest of the traits positive heterosis is desirable. In some cases, for the lowland areas negative heterosis might be desirable for plant height in order to shorten days to flowering and physiological maturity as well as to get lodging free flowering, days to maturity days to emergence etc. negative different. Contrariwise, the comparison of parents vs. hybrids. In other

way, positive heterosis for plant height might be desirable for those areas having long maturity period that is in highland and also intermediate sorghum growing areas so as to increase its biomass.

From the resultant 70 crosses, 37 potential hybrids exhibited significant standard heterosis performance over three checks (ESH-1, ESH-4 and Argiti) in desirable direction for grain yield and among these hybrids 31 hybrids shows significant superiority from the better check. There are also differences for standard heterosis (SH) for yield which was calculated based on relative mean (%), better check hybrid means (%). Most of the crosses displayed positive SH of better check heterosis (BCH) for relative to the highest performing check (better check) where the rest crosses exhibited negative BCH (Table 3).

Similarly, for PHT, there was a significant variability level of hybrid vigour among the studied hybrid parents. In this case MPH ranged from 59.4 to -2.5%. Among the hybrids, 69 of them exhibited positive heterosis while the rest 1 hybrid showed negative heterosis. For the case of BPH heterosis ranged from -23.1 to 44.1% and only 8 hybrids show negative heterosis while the rest are positive heterosis and all positive heterosis ranged between 3 and 44.1%. Like this, there also significant difference of standard heterosis which was done based on mean of better check and this ranged from -16.6 to 29.9% where 11 hybrids exhibited negative BCH and the rest 69 hybrids showed positive BCH. The detailed analysis for heterosis regarding MPH, BPH and BCH is listed out in Table 3.

Significantly, negative BCH for DTF was observed in all the crosses with the range of maximum and minimum heterosis of -0.5 for a cross of 107×76 to -19.5% for 106×86, respectively. Thus, it appeared that the earliest male parent 86 had contributed for earliness, while the late male parent 76 contributed for lateness in comparison of better check heterosis. Among nine significant mid parent heterosis for DTF only four hybrids show significant negative mid parent heterosis while five of them are positively significant different from zero. In this case, negative heterosis was preferable whether it was significant or not for better hybrid vigour to flower earlier than of the better check. BPH ranged from -15.9 by a cross of 106 × 90 to 19.3% by the cross 107 x 86 which was the negative values are goodly selected for better hybrid performance to flower early as compare to that of better parent and the highest positive BPH was not preferable in this condition.

In general, lower value negative heterosis was preferable than higher value of positive heterosis for DTF and DTM. In another way higher value of positive heterosis was good for the rest traits such as GY, TSW, NPT, HL, HE, PW and the like. Heterosis in grain yield for male parent 67 x female line 107 was 0 (Zero). This indicates that as there was additive gene action, and this implies also there is no heterosis. For this case F1 hybrid should be the midpoint or average of both parents.

The per se performance of hybrids for grain yield and its components was in general connected to the heterotic effects. This brought out that selection of hybrids either on the basis of per se performance or on the basis of magnitude of heterotic effects would also be unfailing. This can be supported by yield advantage over OPV standard check. In this case 11 hybrid exhibited higher yield advantage over OPV check. These are across of 106 x 94, 106 x 90, 106 x 102, 107 x 99, 107 x 105, 106 x 95, 107 x 79, 106 x 81, 106 x 92, 107 x 81 and 107 x 94 have yield advantage of ranged from 42 to 75%.

Combining ability analysis

Analysis of variance for combining ability

Combining ability analysis was done following the significance value of genotypes from the general analysis of variance table in the fashion of line x Tester analysis method (Table 1). The nature and magnitude of estimates of genetic variance can deliver an idea about the relative role of fixable (explainable or cumulative) and non-fixable (unexplainable) gene effects in the inheritance of traits. This in turn can help us in identifying suitable parents for hybrid breeding as well as type of breeding method. The genetic variances were estimated from the analysis of variances for combining ability for all traits as suggested by Singh and Chaudhary (1977). The variations among the hybrids were further partitioned into genetic components attributable to GCA and SCA based on the method suggested by Kempthorne (1957). Joint analysis of variance over locations for combining ability indicated that variances due to females was significant for all traits except CHL and TSW, thereby revealing significant contribution of females towards combining ability for these traits. Mean squares due to males were also significant for all the studied traits except HE, and this shows the greatness of good parental selection for this hybrid cross formation which was found highly variable and this was an indication of getting good pollinator lines for hybrid breeding program (sorghum hybrid grain production). The single degree of freedom comparison for parent vs. hybrid, which indicates average heterosis, was significant for all the traits except for DTM and TSW; this clearly suggested considerable amount of average heterosis in the hybrids (Table 4). This also reflected the presence of adequate genetic variability in the experimental material. Similar findings has been reported for average heterosis by comparing parent vs. hybrid in single degree of freedom for 50 hybrids derived from ten female and five male sorghum lines (Kumar et al., 2017).

Combining ability analysis indicated significant GCA mean squares for all measured traits except CHL and TSW for females and HE for males. All traits except PHT,

Table 4. Analysis of variance table for combining ability analysis across locations.

Source of variation	DF	GY	DTF	PHT	HE	NGL	DTM	HL	PW	NPT	TSW
Site	2	35.4**	63.0**	47634.7**	5.9**	0.00	54736.3**	199.8**	633.2**	6.2*	466.6**
Rep (Site)	3	0.3**	0.0	767.1	6.2**	10.2**	96.3**	89.1**	80.3**	5.2*	4.4**
Genotypes	106	2.5**	0.3	3616.4**	2.2**	4.1**	47.6**	54.3**	6.3**	15.7**	2.1**
Parents (P)	36	2.4**	110.6**	3554.0**	3.2**	2.5**	38.6**	39.1**	5.8**	22.3**	43.7**
Hybrids (H)	69	2.3**	77.9**	2186.4**	0.4	4.7**	52.6**	27.0**	6.1**	10.7**	1.2**
Female (FM)	1	3.8**	712.4**	6.9*	2.8**	6.2*	327.7*	267.5**	19.7**	13.4**	10.1
Male (M)	34	2.6**	62.5**	11.5**	0.4	5.8**	49.5**	28.9**	7.4**	8.2**	29.4**
FM x M	34	2.1**	74.6**	1.3	0.4	3.5**	47.7**	17.9**	4.5**	13.2**	24.2
Parents vs. hybrids	1	21.0**	701.9**	104521.1**	89.3**	16.3**	24.4	2480.5**	32.4**	121.9**	12.1
Genotypes*Site	212	1.9**	0.3*	552.6**	0.8**	0.0	43.3**	9.3	4.1**	1.2	1.2*
Parent*Site	68	1.8**	4.8	513.9**	1.6**	0.0	18.2**	8.3	4.3	1.1	16.8
Hybrid*Site	138	1.9**	3.1	543.1**	0.3	0.0	56.7**	9.9	4.1	1.2	20.1
FM*Site	2	6.8**	2.2	1.9	1.5*	0.0	23.5	0.3	0.9	0.1	12.1
M*Site	68	2.0**	3.7	1.5**	0.3	0.0	50.3**	10.9	4.1*	1.4	16.6
FM x M*Site	2	1.6**	2.6	1.6**	0.3	0.0	64.1**	9.0	4.2*	0.9	23.8
Parent vs. hybrids*Site	2	3.5*	8.1	2591.6*	0.4	0.0	23.8	6.7	2.8	2.7	2.6
Error	318	0.96	20.8	311.0	0.56	0.96	12.78	7.66	2.87	1.48	15.4
Total	212										

*P< 0.05; ** P< 0.01; DTF = days to flowering; DTM = days to maturity; GY = Grain yield; HE = head exertion; HL = head length; NGL = number of green leaves; NPT = number of productive tillers; PHT = Plant height; PW = panicle width; TSW = thousand seed weight.

HE, SC and TSW for the female * male interaction were significantly different. In other way, those traits with mean squares due to female * male interaction were significant indicates the presence of genetic variability for SCA among the crosses. These observations further support comparisons of parents vs. hybrids for all traits in a single degree of freedom. Comparing parents with hybrids all measured traits except DTM and TSW was significantly hybrids with site is non-significant except GY and PHT.

The interaction of GCA of females (mean square of females) and site was significantly different only for GY and HE. Similarly, the interaction of site with GCA of males and female x male * site interaction is significantly different for only GY, PHT, DTM and PW and this indicates susceptibility for SCA among the hybrids of the testing sites for these studied traits.

Proportional contribution GCA of females and SCA of males and male*female interaction

In general, the proportional contribution of females, males and their interaction to the total variance showed that males played an important role in the traits indicating predominant male lines influence for these traits (Table 5). It also suggests that in the hybrid sorghum breeding more efforts should be paid to the selection of parental lines.

The highest contributions for grain yield were males and the lowest were the female lines. The proportional contribution of females, males and their interactions (Female x Male) to total variances showed that males played an important role in the traits such as GY, PHT, HE, NGL, CHL, DTM, HL, PW, TSW and SC indicating predominant male influence for these traits (Table 5); and the male to female interaction showed important role for only number of productive tillers. The smaller contribution of interactions of the female x male than males, indicating higher estimates of variances due to general combining ability that is additive gene action among the

Table 5. Proportional contribution of females, males and their interactions to total variance.

Trait	Components										
	GY	DTF	PHT	HE	NGL	CHL	DTM	HL	PW	NPT	TSW
Females	2.43	16.07	1.85	10.50	1.97	0.15	9.41	15.50	474.04	2.17	0.87
Males	55.02	38.71	88.90	42.81	60.92	56.12	47.39	51.39	485.52	36.95	57.64
FM x M	42.55	45.31	9.26	46.84	37.21	43.72	43.20	33.07	321.55	60.89	41.52

males used.

Contribution of interactions of female x male was higher than males for DTF, NPT, indicating higher estimates of SCA variances for interaction. For the same case male lines play important role for days to flowering in this cross being female and male interaction contributing more percentage than the rest (Females and males) did. Similarly, contribution of interactions of female x male was higher than female lines for all traits, indicating higher estimates of SCA variances for interaction. Again, the proportional contribution of males was observed to be higher than that of female x male interactions thus lower estimates of SCA variances.

In general, the proportional contribution of males, females and their interaction to the total variance showed that males played an important role towards the traits indicating predominant male lines influence for traits (Table 5). It also suggested that in the sorghum hybrid breeding more efforts should be paid to the selection of male lines.

General combining ability effects (GCA) for females and males inbred lines

The general combining ability effect of females and males for all measured traits were presented in Table 6. Significant positive and negative GCA effects were observed for all traits. For two Females CHL, NGL and TSW were not found significant and PHT not significant for ATX-623 (female line 106) and the rest significant for both females. Among 35 male lines 12 of them were non-significant for GY. Similarly, for DTF both the negative and positive values showed negative values were selected even if the values were non-significant. In this study; 27 male lines were non-significant for their GCA, but, 17 males were negatively combiner and the rest were positively combiner. The results showed negative and significant GCA effects for days to flowering in the female line (106) and this line induce earliness for the cross which it involves. Male parents including (80, 82 and 92), suggesting the contribution of these parents for earliness in crosses they were involved. Meanwhile, positive and significant GCA effects were observed in 75, 76, 78, 79 and 103 for the same trait. These parents greatly induced lateness in their crosses. The former parents were low general combiners (that is, significant and negative GCA effects). Thirty-two of the crosses expressed negative heterosis over mid parent involving at least one of these parents. The male parent, 86, 78, 76 and 85 greatly induced lateness in their crosses. Out of the two crosses involving male line 80, one of them (female line 106 x male line 80) exhibited significantly negative heterosis over mid and better parent. The lowest significant negative GCA effect (-12.1) was exhibited by male line 80, whereas the highest positive GCA (19.7) was observed in male line 86.

GCA for shorter plant stature (-42.6) was exhibited by male line 75 and the longer one (25.76) was exhibited by male line 105. In the case of GY, the range of maximum and minimum GCA was 1.13 and -0.93 respectively. In this case, male line 75 introduce dwarfness (shortness) for the crosses; it was involved and male line 105 was induced for tallness.

The effects of GCA for male lines revealed that 19 male lines were positively combiner and the rest were negatively combiner. The highest positive GCA value was male line 94 followed by male line 81 and 90 and the lowest negative GCA value was exhibited by male line 74, 88 and 86. Moreover, male line 94 induced high yielding gene effect for the crosses in which it was involved while male line 74 induced low yielding gene effect for those crosses made by male line 74. Among the thirty-five male lines, male line 94, 81, 90 and 92 could be selected for good general combiner for high yield.

Plant height is one of the determinant factors of yield and now a days breeding is to improve grain yield in the same way with biomass. On the contrarily plant height and grain yield is negatively correlated. That means both traits can't improve at the same time. In this study, 12 males were negatively combiner for plant height and the rest 23 males were positively combiner. The 12 male lines could be selected for reducing plant height while the 23 male lines could be selected for increasing plant height. The highest positive GCA

Table 6. GCA effects of males and females.

No.	CHL	DTF	GY	HE	HL	NGL	NH	NPT	PHT	PW	TSW
Females											
106	-0.08^{ns}	-1.34**	0.1**	0.08**	-0.82**	0.12^{ns}	1.68*	0.19*	2.25^{ns}	-0.24*	-0.15^{ns}
107	0.09^{ns}	1.37**	-0.1**	-0.08**	0.78**	-0.12^{ns}	-1.8*	-0.19*	-2.49*	0.24*	0.22^{ns}
SE	0.27	0.31	0.02	0.02	0.19	0.07	0.78	0.08	1.23	0.12	0.27
Males											
71	-0.57^{ns}	1.41^{ns}	-0.39**	-0.12^{ns}	0.69^{ns}	-0.51^{ns}	2.55^{ns}	0.47^{ns}	9.71^{ns}	-0.94^{ns}	0.68^{ns}
72	-1.95^{ns}	-1.18^{ns}	-0.22**	-0.16*	-1.59*	-0.75**	1.05^{ns}	-0.29^{ns}	-6.95^{ns}	-1.6**	0.42^{ns}
73	-0.75^{ns}	0.59^{ns}	-0.43**	0.34**	0.99^{ns}	0.5^{ns}	-2.38^{ns}	-0.91*	-28.2**	0.28^{ns}	-0.9^{ns}
74	0.66^{ns}	-0.17^{ns}	-0.93**	0.17*	1.85*	-1.73**	1.07^{ns}	0.16^{ns}	8.38^{ns}	-0.59^{ns}	1.85^{ns}
75	2.61*	3.07*	-0.49**	-0.11^{ns}	3.56**	0.21^{ns}	-3.15^{ns}	-0.47^{ns}	-42.6**	-0.11^{ns}	-0.93^{ns}
76	-0.54^{ns}	4.68**	0.01^{ns}	-0.14*	-0.51^{ns}	0.49^{ns}	-2.65^{ns}	-0.1^{ns}	12.81*	0.58^{ns}	1.01^{ns}
77	-1.5^{ns}	0.42^{ns}	-0.72**	-0.17*	1.05^{ns}	-0.47^{ns}	-0.51^{ns}	0.39^{ns}	4.05^{ns}	-0.74^{ns}	-2.38*
78	-0.67^{ns}	2.96*	-0.16*	0.01^{ns}	-0.03^{ns}	-0.28^{ns}	2.25^{ns}	-0.02^{ns}	-2.83^{ns}	-0.3^{ns}	0.72^{ns}
79	1.73^{ns}	3.84**	0.12^{ns}	0.12^{ns}	-0.62^{ns}	0.75**	2.5^{ns}	-0.89*	7.41^{ns}	0.55^{ns}	-1.35^{ns}
80	-3.23**	-6.01**	0.05^{ns}	-0.14*	2.69**	-1.01**	4.69^{ns}	1.08**	-40.6**	-1.46**	-0.76^{ns}
81	-0.98^{ns}	-0.83^{ns}	0.7**	-0.17*	-1.11^{ns}	0.79**	6.98*	-0.62^{ns}	16.03**	0.2^{ns}	2.11^{ns}
82	-1.3^{ns}	-2.75*	-0.25**	0.13^{ns}	0.26^{ns}	-1.01**	-0.28^{ns}	0.29^{ns}	17.23**	-0.93^{ns}	1.98^{ns}
83	1.3^{ns}	0.55^{ns}	0.26**	0.01^{ns}	-0.55^{ns}	-0.52^{ns}	1.94^{ns}	0.13^{ns}	0.52^{ns}	0.65^{ns}	-0.66^{ns}
84	2.94*	1.55^{ns}	0.12^{ns}	0.02^{ns}	-0.68^{ns}	-0.01^{ns}	2.14^{ns}	0.75*	18.01**	0.44^{ns}	-2.15^{ns}
85	0.82^{ns}	0^{ns}	0.04^{ns}	0.16*	0.55^{ns}	-0.01^{ns}	1.29^{ns}	-0.64^{ns}	13.52**	0.68^{ns}	0.46^{ns}
86	2.3*	-2.21^{ns}	-0.82**	0.21**	-1.23^{ns}	0^{ns}	-8.86**	1.24**	-5.82^{ns}	1.27**	2.51*
87	-1.28^{ns}	1.42^{ns}	-0.13^{ns}	0.26**	-3.04**	0.47^{ns}	-3.23^{ns}	-0.21^{ns}	1.2^{ns}	0.42^{ns}	-1.38^{ns}
88	0^{ns}	1.65^{ns}	-0.85**	-0.15*	2.26**	-0.97**	-2.32^{ns}	0.02^{ns}	5.78^{ns}	-0.44^{ns}	-1.58^{ns}
89	0.6^{ns}	-1.48^{ns}	-0.17*	-0.08^{ns}	-0.44^{ns}	0.74**	-2.44^{ns}	-0.74*	2.79^{ns}	0.51^{ns}	0.58^{ns}
90	-0.69^{ns}	-2.34^{ns}	0.68**	-0.19**	0.05^{ns}	0.23^{ns}	-3.03^{ns}	-0.8*	15.25**	0.51^{ns}	0.06^{ns}
91	0.22^{ns}	-1.1^{ns}	-0.11^{ns}	0^{ns}	-0.99^{ns}	-0.26^{ns}	1.7^{ns}	0.18^{ns}	-6.27^{ns}	0.08^{ns}	-1.48^{ns}
92	1.85^{ns}	-4.42**	0.6**	0.03^{ns}	-0.41^{ns}	0.76**	0.82^{ns}	-0.27^{ns}	12.41*	0.44^{ns}	1.13^{ns}
93	0.44^{ns}	0.5^{ns}	0.05^{ns}	-0.09^{ns}	1.69*	1.74**	-2.77^{ns}	0.55^{ns}	-33.2**	-0.8^{ns}	0.48^{ns}
94	2.25^{ns}	-1.98^{ns}	1.13**	-0.15*	-2.1**	-0.51^{ns}	4.66^{ns}	0.62^{ns}	13.39**	-0.2^{ns}	2.04^{ns}
95	1.54^{ns}	1.01^{ns}	0.07^{ns}	-0.17*	-1.02^{ns}	0.25^{ns}	-5.1^{ns}	-0.19^{ns}	8.9^{ns}	2.06**	-0.14^{ns}
96	1.37^{ns}	-0.08^{ns}	0.29**	0.27**	-0.18^{ns}	0.74**	-2.02^{ns}	1.86**	-25.9**	0.16^{ns}	1.11^{ns}
97	-1.54^{ns}	-0.93^{ns}	-0.09^{ns}	0.09^{ns}	-1.23^{ns}	0.45^{ns}	-1.95^{ns}	0.06^{ns}	-16.4**	0.14^{ns}	-1.18^{ns}
98	0.68^{ns}	-2.21^{ns}	-0.38**	-0.18*	-0.02^{ns}	-0.73*	-0.8^{ns}	-0.16^{ns}	13.68**	-0.5^{ns}	-1.1^{ns}
99	0.13^{ns}	-1.28^{ns}	0.41**	-0.18**	-0.91^{ns}	0.28^{ns}	0.38^{ns}	-2.17**	6.33^{ns}	1.1*	1.24^{ns}
100	-0.27^{ns}	2.52^{ns}	0.14^{ns}	-0.1^{ns}	-2.13**	0.25^{ns}	-7.46*	-1.51**	3.87^{ns}	-0.34^{ns}	1.59^{ns}
101	-1.14ns	-0.35ns	0.14*	0.35**	-1.22ns	0.79**	-2.69ns	-0.89*	14.61**	-0.41ns	-0.52ns

Table 6. Contd.

102	-0.41^{ns}	-0.61^{ns}	0.49**	-0.16*	2.74**	-0.54^{ns}	0.28^{ns}	0.68^{ns}	-30.4**	-0.51^{ns}	3.75**
103	0.62^{ns}	3.37**	-0.06^{ns}	0.37**	-1.14^{ns}	0.22^{ns}	1.91^{ns}	0.74*	-15.9**	-0.01^{ns}	-1.96^{ns}
104	-3.91**	1.47^{ns}	0.32**	-0.06^{ns}	1.96*	-0.26^{ns}	3.77^{ns}	0.49^{ns}	19.45**	0.02^{ns}	-2.19^{ns}
105	-1.18^{ns}	-0.5^{ns}	0.56**	-0.03^{ns}	0.25^{ns}	-0.01^{ns}	9.53**	1.15**	25.76**	-0.12^{ns}	-1.87^{ns}
SE	1.14	1.3	0.07	0.07	0.8	0.28	3.25	0.35	5.14	0.49	1.15

* P< 0.05; ** P< 0.01; CHL = chlorophyll content; DTF = days to flowering; DTM = days to maturity; GY = Grain yield; HE = head exertion; HL = head length; NGL = number of green leaves; NH = number of heads; NPT = number of productive tillers; PAS = plant aspect; PHT = Plant height; PW = panicle width; SC = stand count; STG = stay green score; TSW = thousand seed weight; SE= Standard Error.

effects were contributed by male lines 105, 104, 84 and 82 respectively. Even though, male lines 75, 80, 93 and 102 were the lowest negative combiner for plant height. Among the highest positive combiner male lines, 81, 102, 99, 94, 90, 105, 104 and 84 were positive significant combiner for grain yield and male line 72, 71, 78, 82, 89 and 91 were significant negative combiner for grain yield.

For the increment performance of hybrids compared to their inbred parents plant height is one of the driving factors for the wider utilization in sorghum production (Mindaye, 2015). The plant height is much better for the need of biomass especially in forage breeding. 20 inbred male parents were significant for the plant height and the rest were not significant, particularly, among 12 of them exhibited negatively GCA effects for the hybrid output. These parents with significant positive GCA effects were good combiners in increasing tallness, while those with significant negative GCA effects were good combiners in decreasing plant height and may be desirable in areas of lodging problem is facing.

The results revealed that male parents were grouped under negatively and positively combiner in the case of days to flowering. Effects of general combining ability analysis revealed that for days to 50% flowering, 16 male parents were positively combined, and the rest 19 males were negatively combined. Only 9 male inbred lines exhibited significant combined effects for days to flowering, and this is an indication that, those with significant negative GCA effects were good combiners in decreasing days to flowering that means to exploit hybrids for their early flowering and those with positively combined effects were better for hybrid exploitation of increasing photo period before flowering. Moreover, parents with positively combined effects greatly induced lateness in their crosses and those with negative effects induce earliness to their respective F1 hybrids.

Panicle length and panicle width were positively correlated with grain yield increment. Male parents exhibited positive combined effects and the rest 20 male lines were negatively combined. So, improving panicle width through heterosis can help to exploit the hybrid grain yield.

In order to get high yielder genetic material for sorghum, panicle width should have wide diameter (width), and among the current study genetic materials, 19 male parents were found positively combined with the respective female lines and the remaining 16 males showed negative general combining ability effects. Male line 95, 86, 99 and 85 could be selected for good positive combiner for wide panicle width. Contrarily, male lines 72, 80, 71 and 82 were the narrowest and poor combiner to increase panicle width for better yield improvement.

Specific combining ability (SCA) effects

The results for SCA estimates detected both negative and positive values for a male line crossed with the two females with equal SCA values in magnitude and opposite in direction but varied among traits that were studied. This may be due to the two females, which had equal combining ability in magnitude but opposite in direction. Similar work was reported in the case of maize using 16 female lines and 2 male lines and the SCA effects was equal in magnitude but opposite in sign (Ejigu et al., 2017).

The estimate of SCA effects for the 70 different hybrids in respect of the traits studied are presented in Table 7. Hybrids evaluated in this study revealed considerable variation in SCA effects in all yield and yield-related traits. It was observed that some crosses involved good general combined parents which produced hybrids, with poor specific combining ability for a given trait example yield, indicating parents with high GCA effects might not always give hybrids with high SCA effects. The possible explanation is that both females used in the hybrid may have the

Table 7. SCA effects for male and female interaction (hybrids) of across sites.

Female	Male	DTF	GY	HE	NGL	NPT	PHT	PW	STG
106	71	5.03**	-0.44**	-0.05^{ns}	0.38^{ns}	-2.83**	-5.29^{ns}	0.27^{ns}	0.16^{ns}
106	72	4.87**	-0.1^{ns}	-0.11^{ns}	1.13**	0.48^{ns}	-3.41^{ns}	0.1^{ns}	-0.62**
106	73	0.99^{ns}	-0.05^{ns}	0.41**	-0.16^{ns}	0.45^{ns}	16.48*	0.46^{ns}	0.29*
106	74	2^{ns}	0.12^{ns}	0.28**	0.14^{ns}	-0.66^{ns}	-6.43^{ns}	0.89^{ns}	-0.01^{ns}
106	75	2.45^{ns}	-0.52**	0^{ns}	-0.84*	1.59**	1.26^{ns}	0.63^{ns}	-0.49**
106	76	-0.15^{ns}	-0.29**	-0.06^{ns}	0.34^{ns}	0.36^{ns}	8.44^{ns}	-0.28^{ns}	-0.18^{ns}
106	77	2.48^{ns}	-0.14^{ns}	-0.06^{ns}	-0.13^{ns}	1.73**	-2.71^{ns}	-0.43^{ns}	-0.02^{ns}
106	78	-1.55^{ns}	0.11^{ns}	0.13^{ns}	0.11^{ns}	-0.58^{ns}	-4.67^{ns}	0.02^{ns}	0.27*
106	79	2.48^{ns}	-0.82**	0.16^{ns}	0.15^{ns}	0.37^{ns}	-0.45^{ns}	-0.58^{ns}	-0.28*
106	80	-0.12^{ns}	-0.11^{ns}	-0.08^{ns}	-0.09^{ns}	-0.67^{ns}	3.36^{ns}	0.34^{ns}	0.02^{ns}
106	81	0.15^{ns}	-0.08^{ns}	-0.07^{ns}	-0.33^{ns}	-1.71**	3.36^{ns}	-0.35^{ns}	0.62**
106	82	0.77^{ns}	-0.02^{ns}	0.26*	1.35**	0.1^{ns}	-1.7^{ns}	0^{ns}	0.24^{ns}
106	83	-3.07^{ns}	0.26*	-0.2*	0.39^{ns}	-1.04*	0.46^{ns}	0.47^{ns}	-0.53**
106	84	1.48^{ns}	-0.37**	0.08^{ns}	-0.62^{ns}	0.12^{ns}	-0.94^{ns}	-0.31^{ns}	-0.08^{ns}
106	85	-2.8^{ns}	0.09^{ns}	0.24*	0.34^{ns}	0.18^{ns}	-6.92^{ns}	-0.34^{ns}	0.45**
106	86	-5.99**	0.51**	0.25*	-0.13^{ns}	0.78^{ns}	-9.75^{ns}	-1.84**	-0.02^{ns}
106	87	1^{ns}	-0.11^{ns}	0.29**	0.41^{ns}	1.53**	-3.04^{ns}	-0.19^{ns}	0.23^{ns}
106	88	2.53^{ns}	0.17^{ns}	-0.1^{ns}	0.38^{ns}	-0.6^{ns}	-9.87^{ns}	-0.02^{ns}	0.09^{ns}
106	89	0.45^{ns}	0.17^{ns}	-0.16^{ns}	0.09^{ns}	1.25*	-1.99^{ns}	-0.83^{ns}	-0.03^{ns}
106	90	-1.14^{ns}	0.66**	-0.08^{ns}	0.1^{ns}	-1.32**	2.1^{ns}	0.3^{ns}	0.54**
106	91	-3.62*	0.57**	0.07^{ns}	0.62^{ns}	1.29*	7.27^{ns}	0.34^{ns}	0.21^{ns}
106	92	0.98^{ns}	0.02^{ns}	0.05^{ns}	0.15^{ns}	-0.74^{ns}	-2.83^{ns}	0.09^{ns}	-0.08^{ns}
106	93	3.49^{ns}	0.3**	-0.18^{ns}	-0.41^{ns}	0.31^{ns}	-2.1^{ns}	-0.05^{ns}	-0.6**
106	94	-3.08^{ns}	0.32**	-0.12^{ns}	0.4^{ns}	-1.64**	-1.86^{ns}	-0.21^{ns}	-0.08^{ns}
106	95	-2.36^{ns}	0.79**	-0.11^{ns}	0.64^{ns}	0.56^{ns}	5.97^{ns}	1.85**	-0.09^{ns}
106	96	-0.12^{ns}	-0.19^{ns}	-0.32**	-0.85*	0.28^{ns}	14.08^{ns}	0.49^{ns}	0.08^{ns}
106	97	-2.13^{ns}	0.19^{ns}	0.21*	-0.61^{ns}	0.29^{ns}	-0.89^{ns}	-0.08^{ns}	-0.08^{ns}
106	98	-2.25^{ns}	0.04^{ns}	-0.08^{ns}	0.09^{ns}	-0.25^{ns}	-4.07^{ns}	-0.19^{ns}	-0.1^{ns}
106	99	0.99^{ns}	-0.94**	-0.09^{ns}	-0.84*	-0.11^{ns}	-1.59^{ns}	-0.32^{ns}	0.14^{ns}
106	100	-1.54^{ns}	0.02^{ns}	-0.12^{ns}	-0.38^{ns}	0.47^{ns}	3.3^{ns}	0.44^{ns}	0.1^{ns}
106	101	-2.06^{ns}	0.28**	0.26**	0.15^{ns}	-0.86^{ns}	5.25^{ns}	0.05^{ns}	-0.25*
106	102	-2.37^{ns}	0.82**	-0.13^{ns}	-0.13^{ns}	-0.61^{ns}	5.13^{ns}	0.66^{ns}	0.62**
106	103	0.05^{ns}	-0.25*	-0.23*	-0.38^{ns}	-0.03^{ns}	1.78^{ns}	-0.18^{ns}	0^{ns}
106	104	0.93^{ns}	-0.28**	-0.15^{ns}	-0.92*	-0.24^{ns}	2.74^{ns}	-0.66^{ns}	0.01^{ns}
106	105	0.7^{ns}	-0.74**	-0.17^{ns}	-0.62^{ns}	1.74**	-6.34^{ns}	-0.61^{ns}	-0.26*
107	71	-5.06**	0.44**	0.05^{ns}	-0.39^{ns}	2.83**	5.53^{ns}	-0.27^{ns}	-0.14^{ns}

Table 7. Contd.

107	72	-4.9**	0.1^{ns}	0.11^{ns}	-1.13**	-0.48^{ns}	3.64^{ns}	-0.1^{ns}	0.63**
107	73	-1.02^{ns}	0.05^{ns}	-0.41**	0.15^{ns}	-0.45^{ns}	-16.24*	-0.47^{ns}	-0.27*
107	74	-2.03^{ns}	-0.12^{ns}	-0.28**	-0.14^{ns}	0.66^{ns}	6.66^{ns}	-0.9^{ns}	0.02^{ns}
107	75	-2.48^{ns}	0.52**	0^{ns}	0.84*	-1.59**	-1.02^{ns}	-0.64^{ns}	0.5**
107	76	0.12^{ns}	0.29**	0.06^{ns}	-0.34^{ns}	-0.36^{ns}	-8.2^{ns}	0.28^{ns}	0.2^{ns}
107	77	-2.51^{ns}	0.14^{ns}	0.06^{ns}	0.13^{ns}	-1.73**	2.94^{ns}	0.43^{ns}	0.04^{ns}
107	78	1.51^{ns}	-0.11^{ns}	-0.13^{ns}	-0.11^{ns}	0.59^{ns}	4.91^{ns}	-0.02^{ns}	-0.26*
107	79	-2.51^{ns}	0.82**	-0.16^{ns}	-0.16^{ns}	-0.37^{ns}	0.68^{ns}	0.57^{ns}	0.29*
107	80	0.09^{ns}	0.11^{ns}	0.08^{ns}	0.09^{ns}	0.67^{ns}	-3.13^{ns}	-0.34^{ns}	0^{ns}
107	81	-0.18^{ns}	0.08^{ns}	0.08^{ns}	0.33^{ns}	1.71**	-3.12^{ns}	0.35^{ns}	-0.61**
107	82	-0.8^{ns}	0.02^{ns}	-0.26*	-1.36**	-0.1^{ns}	1.94^{ns}	0^{ns}	-0.22^{ns}
107	83	3.04^{ns}	-0.26*	0.2*	-0.4^{ns}	1.04*	-0.23^{ns}	-0.48^{ns}	0.54**
107	84	-1.51^{ns}	0.37**	-0.08^{ns}	0.61^{ns}	-0.12^{ns}	1.18^{ns}	0.31^{ns}	0.09^{ns}
107	85	2.77^{ns}	-0.09^{ns}	-0.24*	-0.34^{ns}	-0.18^{ns}	7.16^{ns}	0.33^{ns}	-0.43**
107	86	5.96**	-0.51**	-0.25*	0.12^{ns}	-0.78^{ns}	9.99^{ns}	1.84**	0.04^{ns}
107	87	-1.03^{ns}	0.11^{ns}	-0.29**	-0.41^{ns}	-1.53**	3.27^{ns}	0.19^{ns}	-0.21^{ns}
107	88	-2.57^{ns}	-0.17^{ns}	0.1^{ns}	-0.38^{ns}	0.6^{ns}	10.1^{ns}	0.01^{ns}	-0.08^{ns}
107	89	-0.48^{ns}	-0.17^{ns}	0.16^{ns}	-0.1^{ns}	-1.25*	2.22^{ns}	0.82^{ns}	0.04^{ns}
107	90	1.11^{ns}	-0.66**	0.08^{ns}	-0.1^{ns}	1.32**	-1.86^{ns}	-0.31^{ns}	-0.53**
107	91	3.59^{ns}	-0.57**	-0.07^{ns}	-0.63^{ns}	-1.29*	-7.03^{ns}	-0.35^{ns}	-0.2^{ns}
107	92	-1.01^{ns}	-0.02^{ns}	-0.05^{ns}	-0.16^{ns}	0.74^{ns}	3.07^{ns}	-0.1^{ns}	0.1^{ns}
107	93	-3.52^{ns}	-0.3**	0.18^{ns}	0.4^{ns}	-0.31^{ns}	2.33^{ns}	0.05^{ns}	0.62**
107	94	3.05^{ns}	-0.32**	0.12^{ns}	-0.41^{ns}	1.64**	2.1^{ns}	0.21^{ns}	0.09^{ns}
107	95	2.32^{ns}	-0.79**	0.11^{ns}	-0.65^{ns}	-0.56^{ns}	-5.74^{ns}	-1.86**	0.1^{ns}
107	96	0.09^{ns}	0.19^{ns}	0.32**	0.85*	-0.28^{ns}	-13.84^{ns}	-0.5^{ns}	-0.07^{ns}
107	97	2.09^{ns}	-0.19^{ns}	-0.21*	0.61^{ns}	-0.29^{ns}	1.13^{ns}	0.07^{ns}	0.1^{ns}
107	98	2.22^{ns}	-0.04^{ns}	0.08^{ns}	-0.1^{ns}	0.25^{ns}	4.3^{ns}	0.18^{ns}	0.11^{ns}
107	99	-1.02^{ns}	0.94**	0.09^{ns}	0.83*	0.11^{ns}	1.83^{ns}	0.31^{ns}	-0.13^{ns}
107	100	1.51^{ns}	-0.02^{ns}	0.12^{ns}	0.38^{ns}	-0.47^{ns}	-3.06^{ns}	-0.45^{ns}	-0.08^{ns}
107	101	2.03^{ns}	-0.28**	-0.26**	-0.15^{ns}	0.86^{ns}	-5.02^{ns}	-0.05^{ns}	0.27*
107	102	2.34^{ns}	-0.82**	0.14^{ns}	0.12^{ns}	0.61^{ns}	-4.9^{ns}	-0.67^{ns}	-0.6**
107	103	-0.08^{ns}	0.25*	0.23*	0.38^{ns}	0.03^{ns}	-1.54^{ns}	0.18^{ns}	0.02^{ns}
107	104	-0.96^{ns}	0.28**	0.15^{ns}	0.92*	0.24^{ns}	-2.5^{ns}	0.66^{ns}	0.01^{ns}
107	105	-0.73^{ns}	0.74**	0.17^{ns}	0.62^{ns}	-1.74**	6.58^{ns}	0.6^{ns}	0.27*
SE		1.84	0.1	0.1	0.4	0.5	7.27	0.69	0.13

* P< 0.05; ** P< 0.01; CHL = chlorophyll content; DTF = days to flowering; GY = Grain yield; HE = head exertion; HI = harvest index; HL = head length; NGL = number of green leaves; NPT = number of productive tiller; PAS = plant aspect; PHT = Plant height; PW = panicle width; SC = stand count; STG = stay green score; TSW = thousand seed weight.

same gene controlling effect to the trait(s) studied and not able to take advantage of any additive gene action. Regarding days to flowering, only six hybrid combinations were found highly significant (p<0.01) and one hybrid cross was found significant (p<0.05) probability level. These male parents induced earliness for the crosses where they were involved. But, among these significant hybrids for SCA effects only four (106 x 86, 106 x 91, 107 x 71 and 107 x 72) hybrids were negatively combiner and the rest were positively combiner for this trait.

For days to maturity, only five cross combinations were found to be significant and among these only two were found highly significant (p<0.01) and three of them were significant (p<0.05) probability level. SCA estimates for days to 50% flowering and days to physiological maturity showed both negative and positive SCA effects. Negative SCA estimates for these traits indicated that the crosses took fewer days to 50% flowering and physiological maturity. On the contrary, crosses, which had positive SCA estimates for days to 50% flowering and physiological maturity indicated hybrids with delay in days to 50% flowering and maturity. The current investigation was similar to previous reports in maize using 16 female lines with 2 male lines (Ejigu et al., 2017).

Crosses of female 106 and 107 with male 71, 75, 76, 79, 84, 86, 90, 91, 93, 94, 95, 99, 101, 102, 104 and 105 was highly significant (p<0.01), followed by cross of the same female with male lines 83 and 103 which showed significant at (p<0.05) probability level for grain yield (GY). The range was from -0.94 to 0.94 for cross of male line 99 with 106 and 107 female lines, respectively.

For grain yield among all 70 crosses, a cross of same gene controlling effect to the trait(s) studied and not able to take advantage of any additive gene action. Regarding days to flowering, only six hybrid combinations were found highly significant (p<0.01) and one hybrid cross was found significant (p<0.05) probability level. These male parents induced earliness for the crosses where they were involved. But, among these significant hybrids for SCA effects only four (106 x 86, 106 x 91, 107 x 71 and 107 x 72) hybrids were negatively combiner and the rest were positively combiner for this trait.

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For grain yield among all 70 crosses, a cross of 107x99, 106x102, 107x79, 106x95, 107x105, 106x90, 106x91, 107x75 and 106x86 revealed the highest positive and highly significant SCA effects. On the other hand, crosses such as, 107x77, 106x88, 106x89 and other few have non-significant positive SCA effects. These positive significant and non-significant SAC affected parents indicates that inbred lines involved in these crosses were genetically divergent, and hence could be regarded to be from different heterotic groups. Cross of 106 x 75, 107 x 91, 107 x 90, 106 x 105, 107x95, 106x79, 107x102, 106x99 and some of others showed lowest significant negative SCA effects for this trait, indicating that these crosses were poor in specific combiners for grain yield. Among all crosses that showed the highest significant positive SCA effects, a cross of 107 x 79 and 107 x 75 were a combination of poor female with poor male line in their respected GCA effects. On the contrary, only 2 hybrids 106 x 102 and 106 x 90 were found from a cross combination of good general combiner of female and male. The rest were a cross of poor and good parental general combiner. This showed that, the crosses performed better than what would be expected from the GCA effects of their respective parents. Therefore, these crosses could be selected for their specific combining ability for higher grain yield. When high yielding specific combinations are desired, especially in hybrid sorghum development, SCA effects could help in the selection of parental material for maximum exploitation of heterosis.

Crosses with positive and higher SCA values are desirable for the improvement of productivity of sorghum hybrid grain yield by exploiting maximum heterosis. On the contrary, crosses with negative SCA values are undesirable for sorghum grain yield. However, to get the best SCA, results may not necessarily be from crosses between two good general combiners. From this study, it was found that the combination of a parent with negative and positive GCA value resulted in a hybrid with highly significant positive SCA values in some cases. For example, the combination of a parent with negative GCA female parent and positive GCA male parents gave positive SCA values. Such as across of female line 107 (-0.1) with male line 76 (0.01), male 79 (0.12), male line 84 (0.41), male line 99 (0.41), male line 104 (0.32) and male line 105 (0.56), resulted in positive SCA values of 0.29, 0.82, 0.37, 0.94, 0.28 and 0.74 respectively. The combination of parents with positive and negative GCA values resulting in positive SCA values is also reported from previous investigators (Ejigu, 2017). On the other hand, there were combinations of both parents with positive GCA values, which, resulted in hybrids with negative SCA values, such as female line 106 x male line 76, female line 106 x male line 80, female line 106 x male

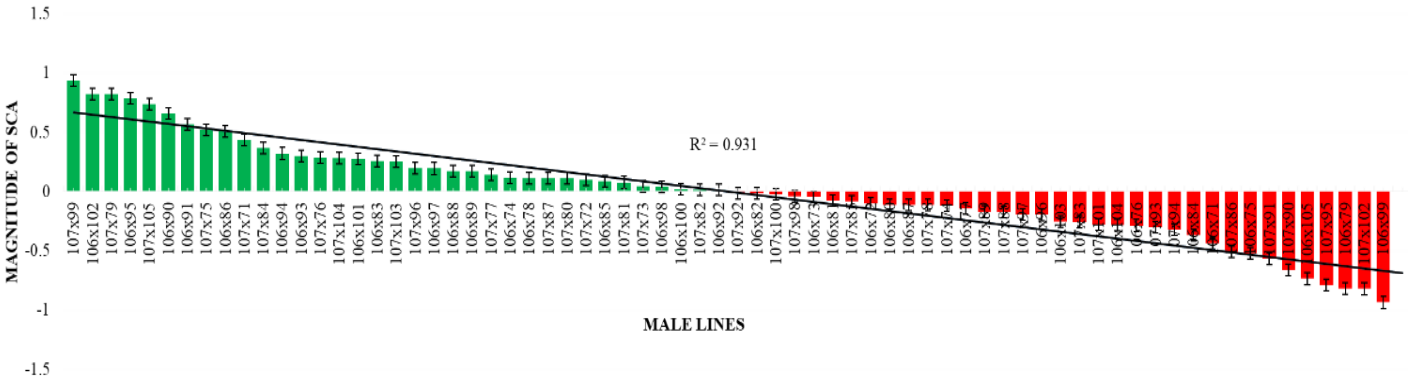

Figure 1. SCA effects for GY across locations of hybrid combinations.

line 81, female line 106 x male line 84, female line 106 x male line 96 and female line 106 x male line 99, female line 106 x male line 104 and female line 106 x Male line 105. The reverse is also true when two parents with negative GCA values were crossed and gave hybrids with positive SCA values such as across of female line 107 with male lines 71, 72, 73, 75, 77, 82, 87 and 103. Therefore, from this study it was found out that the good specific combiners for different traits involved parents with high x high, high x low, low x low general combining ability evaluations.

Among the hybrids, 35 showed positive effects for SCA and the rest showed negative SCA effects. Based on their SCA magnitude the three highest ranks from the top list was a cross of female line 107 with male line 99, 106 x 102 and 107 x 79 respectively. On the other hand, there were also three lowest ranked from the 70 hybrids based on their SCA effects. Those crosses were 106 x 79, 107 x 102 and 107 x 99 and these were the negative reverse of the above former mentioned crosses (Figure 1).

SCA of grain yield derived from female line 106 exhibited positive SCA effect when crossed with male lines 74, 78, 83, 85, 86, 88, 89, 90, 91, 92, 93, 94, 95, 97, 98, 100, 101 and 102. However, hybrid derived from female line 107 exhibited negative SCA effect for those above-mentioned male lines but, equal in magnitude with SCA effects of hybrids derived from female lines 106.

Similarly, days to 50% flowering and plant height also exhibited the same SCA effects. The negative SCA effect is preferred for early maturing traits and this happens due to the rule of the summation of SCA effect is zero since two female lines were used to derive the hybrids.

Gene effects and estimates of variance components for combining ability

*Variances of male, females and their interaction (female * male)*

Estimates of variance indicated the higher SCA variances than the GCA variances for all the studied traits show that predominance of non-additive gene action in the inheritance of these traits and vice versa. The variance of females was found smaller than that of the male lines for all studied traits except NPT which was higher for females than males. The total variance components of parents were contributed by the male lines for cross (hybrid) variances. That is the total hybrid (male * female interaction) variances is its due to male's variation. So, it can be said that selection for male parents for this hybrid production was successful and the next parental selection for hybrid sorghum grain production should take an account of parents for their variance which can put contribution towards the hybrid production (Table 8).

Variances of combining ability effect

Estimates of gca variance (σ2gca), SCA variance(σ2sca) and unity ratio (Table 8). The estimates of variance due to combining ability revealed that σ2gca was lower than σ2sca for all the traits except plant height and number of heads. However, the ratio of σ2gca / σ2sca revealed the preponderance of dominance gene action for all traits except for plant height and number of heads, where additive gene action was more with σ2gca /σ2sca ratio being more than unity.

Generally, the SCA variances (σ2sca) were higher than GCA variance (σ2gca) for almost all the traits except plant height. The ratio of σ2gca/σ2sca was less than one for almost all the considered traits except plant height indicating prevalence of non-additive gene action (dominance and epistasis). Similar investigation was reported for forage sorghum hybrids (Dehinwal et al., 2017). The magnitude of GCA/SCA variance ratio for plant height was specifically sizable, indicating the predominance of additive gene action; however, the specific effects were also highly significant, suggesting the involvement of non-additive effects in controlling this trait. For days to 50% flowering the preponderance of SCA effects (dominance gene effect) was higher than the

Table 8. Variance components of combining ability analysis and estimates of gene effects.

Variance components (σ^2)	GY	TSW	DTF	PHT	NGL	CHL	DTM	HL	PW
Female variance (σ^2fm)	0.01	0.00	3.60	11.05	0.01	0.00	1.20	1.20	0.08
Male variance ($\sigma^2 m$)	0.05	0.66	0.00	287.28	0.18	0.54	0.32	0.78	0.19
Female x Male variance ($\sigma^2 fm * m$)	0.32	0.74	8.14	13.49	0.39	1.17	4.70	1.60	0.27
Vgca ($\sigma^2 gca$)	0.07	0.86	1.35	386.82	0.25	0.64	1.24	1.85	0.30
Vsca ($\sigma^2 sca$)	1.27	2.96	32.58	53.96	1.55	4.67	18.81	6.42	1.09
Unity variance ($\sigma^2 gca/\sigma^2 sca$)	0.06	0.29	0.04	7.17	0.16	0.14	0.07	0.29	0.28
Additive variance $\sigma^2 A$	0.14	1.72	2.71	773.65	0.49	1.29	2.48	3.70	0.61
Dominance variance $\sigma^2 D$	1.27	2.96	32.58	53.96	1.55	4.67	18.81	6.42	1.09
Degree of dominance ($\sigma^2 D/\sigma^2 A$)	0.11	0.58	0.08	14.34	0.32	0.28	0.13	0.58	0.56

additive gene effects and this indicates that hybrids are earlier than their parental effect. Similar investigation was done for 7 lines and 8 testers of 28 forage hybrids in relation to days to 50% flowering (Mohammed, 2009). For all traits, SCA variances found higher preponderance of additive gene action heterosis breeding will be effective. Contrarily, for those traits such as plant height and number of heads' GCA, variances are higher than SCA variances (Table 3).

The degree of dominance (σ2D/σ2A) was found greater than one for all the traits except plant height, indicating the over dominance behavior of interacting alleles. Since over dominance geneaction is involved for inheritance of grain yield, heterosis breeding would be the most effective approach to improve the trait. The significance of mean square for female x male provides a direct test of significance of dominance variance, σ2D while significance of σ2A is provided by significance of females and males mean squares (Nduwumuremyi et al., 2013) (Table 8). The results revealed that for plant height, stay green score, and other few, the additive genetic effects were more pronounced than non-additive effects, and the general combining ability variance was higher than specific combining ability. This result suggests that the inheritance of these traits was mainly controlled by additive genes and selection of parents should be more important in breeding practice.

Estimates of gene action

Detail estimates of additive variance (σ2A), dominance variance (σ2D) and average degree of dominance (σ2D /σ2A) on the basis of pooled analysis over sites (Table 8). The estimates for components of genetic variance, that is, σ2A and σ2D were computed from the variance of combining ability calculated on the basis of the covariance of half sib and full sib as suggested by Singh and Chaudhary (1977). Both additive and non-additive gene actions are expected to be important in the expression of the studied traits, with the preponderance of additive gene actions for plant height and stay green and non-additive actions for all traits except plant height and stay green (Table 8).

Estimation of components of variance (additive variance from GCA and dominance variance from SCA variance), and subsequent estimation of average degree of dominance (σ2D/σ2A) was more than unity (one) for most of the studied traits during pooled analysis. This further indicates the preponderance of dominance gene action for most of the traits except plant height, number of heads and stand count where additive gene action seems to be more important (Figure 2) and this agrees with similar findings in forage sorghum (Kumar et al., 2017). For those traits like PHT, NH, SC and STG with degree of dominance was found greater than unity (one) in both the additive and non-additive gene actions which were responsible for inheritance of grain yield and its components in sorghum hybrid grain production.

Clustering analysis of experimental genotypes

Estimation of cluster likelihood was calculated based on five independent runs (R software [Version 5.1]) for a variable number of clusters, from K = 2 to 10. K = 3 was chosen due to their low variation of probability values and repetitive clustering. Clustering analysis based on the genetic dissimilarity grouped the 110 sorghum

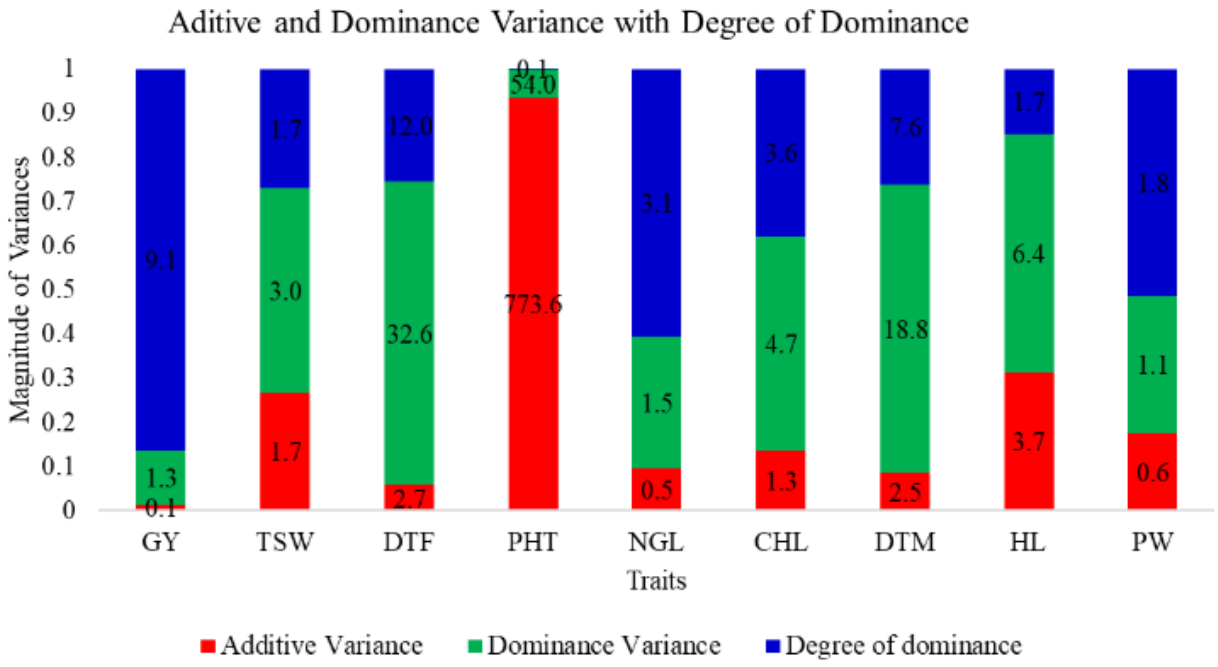

Figure 2. Estimate of gene action for selected traits.

genotypes into 3 distinct clusters. It was observed that cluster I had maximum number of genotypes followed by cluster II and III with 59, 29 and 22 genotypes respectively.

In cluster III, all genotypes are parental genotypes except genotype 45 and 61. In cluster I, most of the grouped genotypes are also parents and some are hybrids (crosses). Both female lines are grouped under cluster III and this indicates that both female lines are similar in their performance. In this case, the dissimilarity range is found between 16.2 to 23.1%. Cluster III had 22 genotypes reflecting narrow genetic diversity amongst them. The narrow genetic diversity may be attributed to similarity in the base material from which they have been evolved. Therefore, it can conclude that parental lines were more variable than their hybrids derived from them since hybrids are concentrated more in cluster I while parents are distributed in clusters II and III (Figure 3).

Heritability and variances of genetic components

The extent to which variation in yield components are responsible for differences in yield among various cultivars, depends on heritable and non-heritable components. While coefficients of variation measure the magnitude of variability present in a population estimates of heritability. The estimates of PCV and GCV were classified as low (< 10%), medium (10-20%), and high (> 20%) (Sivasubramanian and Madhavamenon, 1973). The results revealed considerable phenotypic and genotypic variances among all the genotypes for the traits under consideration for this study. In most traits a large portion of the phenotypic variance was accounted for the genetic component and the contributions of genetic variance to phenotypic variance were found high (Table 9). The estimates of PCV were high for all studied traits ranging from 5.3 of days of flowering to 64.1 of number of productive tillers except for number of productive tillers and number of green leaves which are equal in their GCV and PCV. The PCV values were higher than GCV values for all the traits can reflect the influence of environment on the expression of traits. High PCV values were recorded for GY, PW, NPT and TSW while DTF and NGL were found low. For the case of GCV only for NP is found high value while for GY, PHT, HL, NH and NGL were found medium. All the other traits showed moderate PCV and GCV values. For those traits like grain yield, panicle width, head length, number of productive tiller and number of green leaves in which genotypic variance is higher than error variance, genetic variance is more important, and selection can be done if possible. Similarly, for trait which is GCV is higher indicates that there were low environmental effects on these traits.

Heritability estimates (broad-sense) for yield and its components were done following Singh and Chaudhary (1979). They were categorized using the criteria of Robinson and Comstock (1949): 0-30% = low; 31-60% = moderate; > 60% = high. High heritability estimate was recorded for all traits while for days to maturity was found low and for panicle width, thousand seed weight and number of heads were found moderately heritable. This indicates that these traits are highly heritable, and it would give the best picture of genetic advance to be

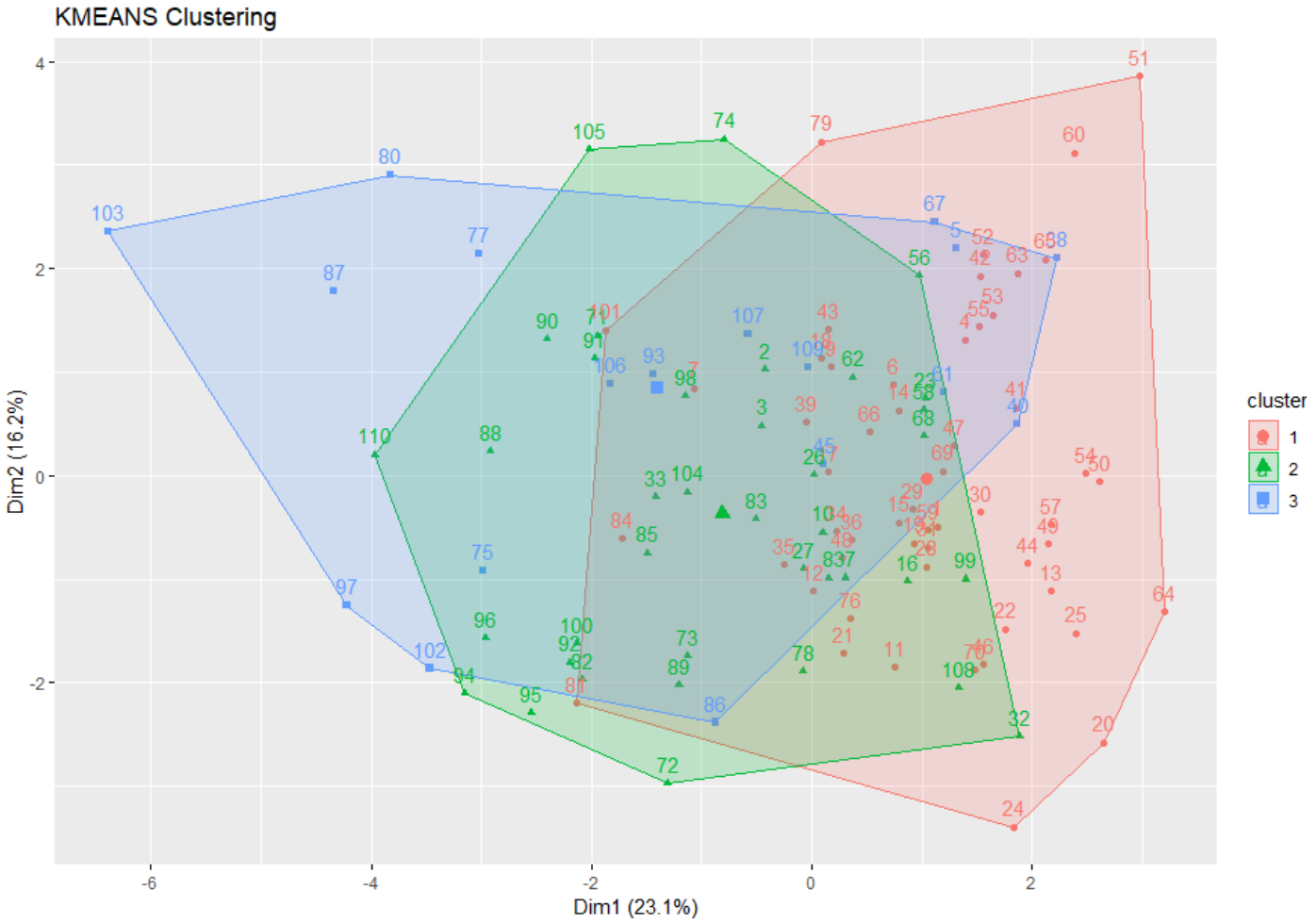

Figure 3. Kmeans clustering of genotypes.

expected from selection.

Heterotic grouping

Two heterotic grouping methods were used to assign male parents into different groups, including SCA effects for grain yield (Pswarayi and Vivek, 2008) and heterotic grouping based on GCA of multiple traits (HGCAMT) method (Badu-Apraku et al., 2013). Dendrogram was constructed for the groupings based on HGCAMT. Using Ward's method based on Euclidean distances obtained from GCA for all traits. Classification by these two methods showed similar but not identical trends. As of two female lines (106 and 107) were used in this cross, the principle of SCA effects method is as follows: Male parents showing negative SCA effects when crossed with 106 and exhibiting positive SCA effects with 107 were classified into heterotic Group A. Male lines showing positive SCA effects with 107 and negative effects with 106 were assigned into heterotic Group B, similar result was reported by Akata et al. (2017) for heterotic grouping of 19 male lines crossed with two female lines into 4 groups based on their SCA effects. The SCA effects of grain yield method classified those sorghum elite lines into two heterotic groups (Table 10). Based on this heterotic group, 16 elite lines were found under group A with negative SCA effects for hybrids derived from female line 106 and the rest 19 elite lines were laid under group B which had positive SCA effects for cross of female line 107. Grouping of inbred lines based on their GCA effects of multiple traits should give a better, more probable and practical heterotic group of the lines since GCA dealings the additive gene effects for each trait. Dendrogram based on HGCAMT method grouped sorghum inbred lines into two heterotic groups (Figure 4). In group I, twenty inbred lines were grouped together and in group II fifteen lines were found. Inbred lines of both groups had negative and positive significant GCA effect for grain yield and flowering time; while almost all group I individuals were lateness lines. The highest positive highly significant five lines were being found under group

Table 9. Estimates of heritability and genetic components.

Component	GY	TSW	DTF	PHT	CHL	DTM	HL	PW	HE
H^2	68.5	41.4	86.8	85.7	71.3	12.5	84	31.3	23.8
h^2	6.8	10.6	5.6	70.5	7.8	6	23.6	16.6	3.5
GCV (%)	12.9	5.4	4.9	13	4.6	0.8	10.5	8	13
PCV (%)	42.5	23.4	5.3	16.5	6.4	13.5	11.4	27.9	17.8

Table 10. Heterotic groups of Ethiopian sorghum elite lines identified by SCA of grain yield.

Male	F106		F107		Heterotic groups	GCA of males
	Mean GY	SCA	Mean GY	SCA		
71	1.94	-0.44**	2.62	0.44**	A	-0.39**
72	2.45	-0.1^{ns}	2.45	0.1^{ns}	A	-0.22**
73	2.29	-0.05^{ns}	2.19	0.05^{ns}	A	-0.43**
74	1.95	0.12^{ns}	1.52	-0.12^{ns}	B	-0.93**
75	1.75	-0.52**	2.6	0.52**	A	-0.49**
76	2.48	-0.29**	2.86	0.29**	A	0.01^{ns}
77	1.9	-0.14^{ns}	1.98	0.14^{ns}	A	-0.72**
78	2.72	0.11^{ns}	2.3	-0.11^{ns}	B	-0.16*
79	2.06	-0.82**	3.51	0.82**	A	0.12^{ns}
80	2.7	-0.11^{ns}	2.73	0.11^{ns}	A	0.05^{ns}
81	3.39	-0.08^{ns}	3.35	0.08^{ns}	A	0.7**
82	2.5	-0.02^{ns}	2.34	0.02^{ns}	A	-0.25**
83	3.28	0.26*	2.58	-0.26*	B	0.26**
84	2.52	-0.37**	3.06	0.37**	A	0.12^{ns}
85	2.88	0.09^{ns}	2.52	-0.09^{ns}	B	0.04^{ns}
86	2.45	0.51**	1.25	-0.51**	B	-0.82**
87	2.52	-0.11^{ns}	2.55	0.11^{ns}	A	-0.13^{ns}
88	2.08	0.17^{ns}	1.55	-0.17^{ns}	B	-0.85**
89	2.77	0.17^{ns}	2.23	-0.17^{ns}	B	-0.17*
90	4.1	0.66**	2.59	-0.66**	B	0.68**
91	3.22	0.57**	1.9	-0.57**	B	-0.11^{ns}
92	3.37	0.02^{ns}	3.15	-0.02^{ns}	B	0.6**
93	3.11	0.3**	2.32	-0.3**	B	0.05^{ns}
94	4.22	0.32**	3.38	-0.32**	B	1.13**
95	3.62	0.79**	1.85	-0.79**	B	0.07^{ns}
96	2.85	-0.19^{ns}	3.05	0.19^{ns}	A	0.29**
97	2.86	0.19^{ns}	2.29	-0.19^{ns}	B	-0.09^{ns}
98	2.42	0.04^{ns}	2.15	-0.04^{ns}	B	-0.38**
99	2.23	-0.94**	3.92	0.94**	B	0.41**
100	2.92	0.02^{ns}	2.68	-0.02^{ns}	B	0.14^{ns}
101	3.18	0.28**	2.43	-0.28**	B	0.14*
102	4.07	0.82**	2.23	-0.82**	B	0.49**
103	2.45	-0.25*	2.77	0.25*	A	-0.06^{ns}
104	2.8	-0.28**	3.17	0.28**	A	0.32**
105	2.58	-0.74**	3.86	0.74**	A	0.56**
Mean	2.8		2.6			

A= males had negative SCA effects with female lines 106; B= males had positive SCA effects with female lines 107.

I while the other lowest found in group II for grain yield. Eight of lines were grouped under group II in both of

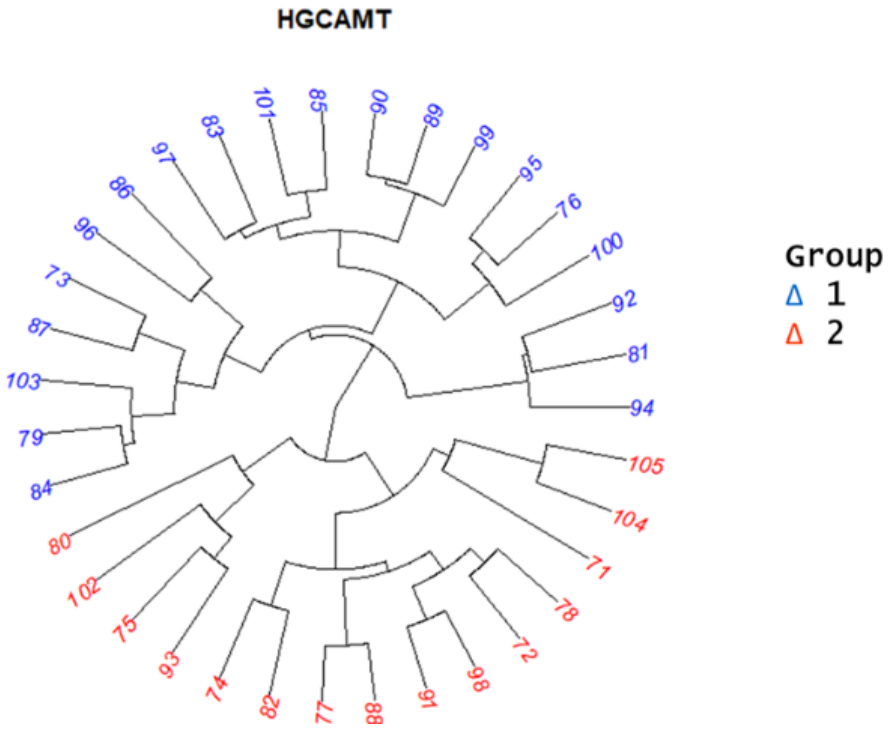

Figure 4. Dendrogram of 35 sorghum male inbred lines constructed from GCA effects of grain yield and other traits (HGCAMT) using Ward's grouping analysis across sites.

grouping methods while other eight lines also were grouped under group I use these two grouping methods. The HGCAMT method provides information related to plant performance and general combining ability of multiple traits and can be supported by correlation pattern between per se performance and general combining ability. Furthermore, while HGCAMT tended to put most of the inbred lines with positive GCA effect for grain yield into group I and the ones with negative values into group II, the SCA method puts most of the inbred lines in to group A.

Examination of the results of the classification of the sorghum inbreds based on the SCA effects of yield and HGCAMT, fifteen inbreds were grouped similarly by both grouping methods (SCA and HGCAMT). It is striking that the assortment of the sorghum inbreds in an earlier study using the SCA method (Akata et al., 2017) and HGCAMT method used by (Badu-Apraku et al., 2013) showed close correspondence with the classification by the HGCAMT and SCA methods used in the present study.

Conclusion

Analysis of variance for all genotypes showed that highly significant difference for all measured traits and this revealed that as there was variability between genotypes included in this investigation. Parents and hybrids discovered that mean squares due to parents for all the traits were found highly significant indicating considerable amount of variability among the parents for various traits. The mean squares due to hybrids were also found highly significant for all the traits except head exertion. Parents vs. hybrids comparisons were found highly significant for all traits except TSW, DTM and STG indicating substantial amount of heterosis among hybrids. This indicates that there is high variability between females and male interaction and by selecting best hybrid combination exploitation of hybrid vigour for commercial use can be done. An examination of per se performance of parents and hybrids with their standard checks revealed that genotypes 24, 20, 32, 64, 70, 25 and 44 among hybrids and among parents, male line 72 and 81 exhibited higher mean performance for grain yield and some of the yield attributing traits.

Male line 79, 96, 94, 102 and 81 had desirable significant GCA effects for grain yield and other agronomic traits and could be used in the breeding program for the development of high yielding hybrids with desirable agronomic performance. The significant male and female (GCA) and hybrids (SCA) mean squares observed for measured traits showed that both additive and non-additive gene actions were important in the inheritance of the traits. This suggests that significant breeding progress could be achieved using both inbreeding and hybridization. Higher magnitude of SCA

variances found for all traits studied depicted the relative predominance of non-additive gene effects in the inheritance of these traits. This result is in agreement with the report done by using two female and nineteen male lines (Akata et al., 2017).

Sufficiently high magnitude of heterosis in desired direction was observed for all the traits and heterobeltiosis was also observed in desired direction for all the traits. Similarly, better check heterosis (economic heterosis regarding better check) also showed relevant hybrid vigour as compare to high (good) performing check among checks. For BCH, the higher estimate of heterosis for grain yield was registered by a cross combination of 106 x 94, 106 x 90 and 106 x 102. 27 crosses showed significant positive heterobeltiosis (BPH) and 19 of the hybrids exhibited significant positive effects of heterosis regarding better check hybrid vigour (higher standard heterosis) for grain yield. The better check heterosis for grain yield was registered by the cross of 106 by 94, 90, 102, 95, 95, 81 and 92 and across of 107 by 99, 105, 79, 81 and 94 in respective of the order in their magnitude of heterosis.

The estimation of general combining ability variances for female lines (σ2fm) were found higher than variances of male lines for traits of DTF, DTM, HL, NH and SC. While general combining ability variance for males (σ2 m) was found higher for all traits except DTF, DTM, HL, NH and SC. On the other hand, specific combining ability (SCA) variances for hybrids were higher than the variances of GCA for all traits except PHT and NH. This indicated the predominance of dominance gene action for most of the traits.

Heritability in this study showed that as there was considerable significant magnitude and ranged from 12.5 for days to physiological maturity to 93% for number of productive tillers which was highly heritable while heritability for days to maturity was highly influenced by environmental effect and it was found as low heritable.

Based on this impact, elite lines used in the present study is grouped under two heterotic groups based on their SCA effects expressed for each of male lines crossed with each of female lines. So, based on their grain yield SCA effect 16 lines were found in group A which has negative GCA effects and the other 19 male lines were laid under group B due to their positive GCA effects. So, for the case of grain yield, always positive effects are preferable. Based on this statement, group two male lines were found to be good positive combiner to boost grain yield in sorghum hybrid program.

This study highlighted hybrid performance, combining ability effects and heterotic responses among Ethiopian elite sorghum lines for important traits studied indicating the existence of favorable alleles for breeding programmes. Specific environment-based improvement may allow good progress rather than over location adaptation. The dominance gene effect was predominant in the inheritance of these traits suggesting direct selection methods. Female parent 106 followed by 107 and male parents 94, 102, 90 were the best combiners for grain yield and other related traits providing opportunity for breeders to improve grain yield under diverse environments. 106 x 94, 106 x 102, 106 x 90, 106 x 81 and 107 x 105 were found to be the best cross combinations for superior hybrids and should be tested extensively in multilocation trials and promoted for adoption and commercialization in dry low land areas of Ethiopia to improve food security. The HGCAMT method was suitable in superior hybrid prediction and the heterotic grouping information could be useful for sorghum breeders using local adapted elite lines to identify best parents for superior hybrids development in Ethiopia.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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