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Full Length Research Paper

Effect of nitrogen fertilizer treatments on duration of senescense, harvest time and yield in some varieties of cowpea (*Vigna unguiculata* (L.) Walp)

Mohammad M.[1]* and Mukhtar F. B.[2]

[1]Department of Biological Sciences, Federal University, Dutse, Jigawa State, Nigeria.
[2]Department of Biological Sciences, Bayero University, Kano, Kano State, Nigeria.

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The effects of liquid nitrogen fertilizer (150% N) and benzyl amino purine (BAP 200 ppm) on senescence, yield and nutrients mobilization in two cowpea varieties were investigated in the rainy and dry seasons. The study was conducted in a screen house at the University of Lagos, Nigeria (6°27"1, 3°45" E). The duration of the study was between August 2012 and January 2013. The experimental design used was completely randomized block design. The four treatments (nitrogen fertilizer, BAP, nitrogen fertilizer and BAP combination in 3:1 ratio and the control) were applied through foliar spray at 3, 6 and 9 weeks after planting and data on time of senescence and yield was collected. The days to 50% senescence, 90% senescence and death of the plants was earlier in IT89KD-288 than in Kanannado, while senescence started earlier in plant treated with liquid nitrogen fertilizer (15%N). For the first harvest, variety IT89KD-288 had higher number of pods (10), number of seeds (13) and length of pods (15.8 cm) than kanannado that had 8.0, 12.0, 14.68 cm, 3.17 and 18.53 for number of pods, number of seeds, length of pods, weight of grains per pod and weight of grains per plant, respectively, while at the second harvest, kanannado had higher number of pods (5), number of seeds (10), length of pods (12.12 cm), weight of grains per pod (2.68) and weight of grains per plant (12.03) than IT89KD-288 that had 4.0, 9.0, 11.31 cm, 2.05 and 11.27 for number of pods, number of seeds, length of pods, weight of grains per pod and weight of grains per plant, respectively. The combined treatment induced significantly greater yield than the other treatments. Yields were higher in the rainy season than dry season. The longer the duration of senescence, the higher the grain yield at the second harvest; therefore, confirming a direct relationship between senescence, harvest time and yield among cowpea varieties.

Key words: Cowpea, benzyl amino purine, nitrogen, kanannado, fertilizer.

INTRODUCTION

Cowpea, *Vigna unguiculata* (L.) walp, subspecies unguiculata is a dicotyledenous spermatophyte belonging to the family fabaceae. The plant is an annual crop grown virtually all over the world. There are several cultivars adapted to different world climatic regions. The crop is widely cultivated in Nigeria for the consumption of its leaves, green pod and grain. The herbage can also be used as green manure and animal feed (Steele, 1976).

Cowpea is an important food grain legume for over 200 million people in the dry savanna of tropical Africa. It is particularly important in West Africa with over 9.3 million metric tonnes of annual production (Oritz, 1998). The grain is a good source of human protein, while the haulms are valuable source of livestock protein (Fatokun, 2015). It is also a source of income for many smallholder farmers in sub-Saharan Africa and contributes to the sustainability of cropping systems and soil fertility improvement in marginal lands through provision of ground cover and plant residue, nitrogen fixation and suppressing weed. However, despite its great importance, grain yield of cowpea crop is low, about 300 kg ha^{-1} (Cardoso et al., 2015; Leite et al., 2015). When compared with many other crops, cowpea has received little attention from plant breeders and large efforts need to be made to break the yield barriers and for cowpea production to keep pace with other crops, especially cereals, its yield potential must be improved (Anonymous, 2014).

In Nigeria, 80% of the cowpea produced mainly as grain is from the savanna zone of the country (FAO, 1999). A wide range of seed yield**s** has been recorded for cowpeas but is generally low. Among factors responsible for the low yields is low soil fertility, as most tropical soils are deficient in essential nutrients, particularly nitrogen and phosphorus (Jones and Wild, 2013). Traditionally, soil fertility in West Africa has been maintained through fallow. However, in Nigeria, intensive cropping is gradually replacing the traditional shifting cultivation that is associated with long fallow and hence low crop yield. The steady decline in food production due to reduced length of fallow on land has prompted farmers to amend soil with different materials (organic and inorganic) to enhance plant growth and increase yield (Adepetu, 2013). It has been suggested that organic manure should be used in place of chemical fertilizer to avoid long-term negative effects of chemical fertilizer on the soil. However, organic manure is usually required in large quantity to sustain crop production and may not be available to the small-scale farmers (Nyathi and Campbell, 1995), hence, the need for inorganic fertilizer. The positive effect of the application of inorganic fertilizers on crop yields and yield improvement have been reported (Carsky and Iwuafor, 2013). Although, cowpea symbiotically fixes nitrogen, plant dependent on symbiotically fixed N may well suffer from temporary N deficiency during the seedling growth once the cotyledonary reserves have been exhausted.

Usually, prior to the onset of symbiotic N fixation, cotyledonary reserves are mobilized during hypocotyl elongation in cowpea and cotyledons are usually shed one or two days from emergence. It has thus been recognized and demonstrated that application of a small quantity of nitrogen fertilizer enhances early vegetative growth (Dart et al., 2007). Burris (2014) stated that nitrogen has a stimulating effect on root activity and rooting pattern of the crop. It has also been reported that available nitrogenous compound allowed seedlings to make a good start before nitrogen fixation has a chance to occur. Other workers have shown that plants given inorganic N during vegetative periods were much larger by the onset of flowering than those dependent on symbiotic N fixation (Minchin et al., 2014). Such plants also had more branches and produced many peduncles resulting in greater number of pods, seeds and significantly larger yields. There are many reported studies on the effects of P application on growth and yield of cowpea (Owolade et al., 2006; Kolawole et al., 2002; Okeleye and Okelana, 2013) there is dearth of information on the effects of N fertilizer on growth and yield of cowpea in Nigeria. However, it has been reported elsewhere that the main limiting nutrients for legume production in the tropics are N and P (Fox and Kang, 2009).

The term senescence is basically derived from the Latin verb *senescere* meaning “to grow old”; generally, the most obvious senescence in plants is foliar senescence (leaf senescence). In fact, the leaf senescence is the last stage of leaf development during which the leaf color changes from green to yellow (Keech et al., 2007). Normally, leaf senescence is initiated by yellowing of the margins of the leaf blade extending towards center of the leaf blade near the midrib, resulting in death of the leaf. Leaf senescence although deteriorative in nature has been recognized as the last phase of the organs development, a highly ordered process regulated by genes known as senescence associated genes (SAGs) (Pruitt, 1983). The leaf when young and mature accumulates nutrients and exports them to growing parts of the plant during senescence

In cowpea, senescence causes substantial reduction in total grain yield because most cowpea plants die after producing the first flush of pods. The reduction in yield is most drastic in the local varieties. Delaying leaf senescence will most probably extend the reproductive period and increase the photosynthetic efficiency of the crop resulting in increased grain yield. The objective of the experiment is to investigate the combined effect of liquid nitrogen fertilizer with cytokinin (BAP) in regulating the onset of senescence in some cowpea varieties and also to determine the combined effect of nitrogen fertilizer and BAP on yield of two cowpea varieties.

MATERIALS AND METHODS

Study area

This study was conducted in the screen house at the University of Lagos located in the south-western part of Nigeria, latitude 6°27"N,

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*Corresponding author. E-mail: maryammohammad35@yahoo.com. Tel: 08023778967.

longitude 3° 45" E and altitude 0 to 41 m (0-135 ft) above sea level with a tropical wet and dry climate of two peaks of rainy seasons, from April to July and between October and November. There is a brief relatively dry spell between August and September and a longer dry season from December to March. Monthly rainfall averages over 400 mm (16 in) between May and July, 200 mm (7.9 in) in August and September and as low as 25 mm (0.98 in) in December. The temperature ranged from a maximum of 37.3°C to a minimum of 13.9°C (World Weather Information Service Lagos, 2012).

Source of seeds

Kanannado, a local cowpea variety and IT 89KD–288, an improved variety which are photosensitive, were collected from IITA, Ibadan. In cowpea, senescence causes substantial reduction in total grain yield because most cowpea plants die after producing the first flush of pods. The reduction in yield is most drastic in the local varieties of which kanannado and IT89KD-288 are among the most affected hence their selection for the experiment.

Kanannado

This is a local variety that originated from Kano in the northern part of Nigeria. It is strongly photosensitive and late maturing, requiring about 80 to 114 days to flower during the rainy season when average minimum and maximum temperatures are between 19.6 and 32.5°C, respectively and day length range from 13.6 to 12.9 h day^{-1}. It has an indeterminate growth habit with lateral branches growing up to 3 m long during the wet season. On the other hand, when such plants are grown where there is no rainfall and day length is short (about 12.2 to 12.7 h day^{-1}) with minimum and maximum temperatures between 15.2 and 30.7°C respectively, their growth becomes stunted due to lack of elongation of the internodes. Branching habit is also affected in this case and the plants assume a rosette appearance. However, maturity occurs early (takes 40 to 45 days from sowing to flowering) (Singh, 1997). They have very dark green leaves and white flowers, pods are non-pigmented, coiled with thin pod walls. Seeds are large, rough and white with brown hilum.

IT89KD–288

IT89KD–288 is a cultivar derived from the cross between IT897F–1772 (Kanannado selection) and IT845–2246–4, like Kanannado, it is strongly photosensitive and late maturing requiring similar number of days to flower during the rainy season. Growth is indeterminate with lateral branches up to 3 m long. Because of its photosensitive nature, there is stunting growth in the dry season when day length is short; however, maturity is early, leaves are dark green, flowers are white, the pods are unpigmented, slightly curved and seeds are large, rough and white. It yields more fodder and grain than Kanannado. It combines resistance to aphids, bruchids and thrips.

Planting

Plastic pots of 250 mm diameter were used in this study. 240 pots were used for the two varieties, 120 pots for each variety. They were filled with fresh sandy loam top soil and watered well for 2 days before planting. The treatments were replicated 8 times in completely randomize block design. The treatments used include benzyl amino purine (BAP) and liquid N fertilizer (Boost Extra). There were also combined treatments of BAP and liquid N fertilizer. The 1st planting was done August 9th, 2012 for the rainy season and the 2nd planting was done January 20th, 2013 for the dry season. Seeds were directly sown in the prepared pots (after surface treatment with fungicide Apron plus) with 2 seeds per pot. They were labeled appropriately using white and green plastic tags according to the applied treatment.

Experimental treatments

Liquid nitrogen fertilizer

The type of liquid fertilizer used was Boost Extra, the composition of this fertilizer includes: nitrogen 20%, phosphate 20%, potassium 20%, magnesium 1.5%, iron EDTA 0.15%, manganese EDTA 0.075%, copper 0.075%, zinc 0.075%, boron 0.0315%, cobalt EDTA 0.0012% and molybdenum 0.0012%. The pH of the solution (10% solution) was 4.0 to 4.5. One hundred millilitres (100 ml) of the liquid fertilizer (Boost Extra) was added to 15 L of water, mixed together and applied to the plants using foliar application technique. The fertilizer was applied at 3, 6 and 9 weeks after planting.

Hormone treatment

The hormone used in this research was BAP at 200 ppm concentration and there were 15 replications per treatment. A foliar spraying technique was used to apply the treatment at three doses: at 3, 6 and 9 weeks after planting. The control plants were sprayed foliarly with water in 3 doses every week.

Combined treatments

The combined treatments comprise of a combination of the hormone and the liquid fertilizer, the liquid fertilizer was applied first and the plants were subsequently sprayed with cytokinin (BAP) at 3:1 ratio of liquid fertilizer and hormone, respectively.

Data collection

Data was collected on senescence, the changes in leaves color were observed visually and the extent of chlorophyll loss was monitored. Days to onset of senescence were counted and recorded for each plant, likewise days to 50% of senescence, days to 90% of senescence and days to complete death of the plant. From the obtained data, the duration of senescence were estimated in order to determine their effects on senescence in the cowpea varieties. Data was also collected on yield as follows:

Number of pods

Number of pods for each treated and control plants were counted and recorded.

Length of pods

The length of each pod of the plants was measured with the aid of a meter rule and a thread and the values were recorded.

Number of grains per pod

The number of grains per pod for each plant was counted and recorded.

Table 1. Mean number of days to onset, progression and duration of senescence in the cowpea varieties, treatments and season.

Treatment	Onset of senescence	50% senescence	90% senescence	TDOP	DOS
Variety					
IT89KD288	64	81	93	102	38
Kanannado	72*	91*	102*	114*	42*
Mean	68	86	98	110	40
LSD (0.05%)	2.41	2.98	2.87	2.17	1.98
Foliar fertilization					
150% N	66	82	96	105	39
200 ppm BAP	77*	97*	106*	120*	42
150% N + 200 ppm BAP	72	96	110	121	48
Control	57	69	81	92	35
Mean	68	86	98	110	41
LSD (0.05%)	5.41	7.43	4.34	3.84	2.82
Season					
Rainy	69*	81*	104*	117*	47*
Dry	67	91	91	102	35
Mean	68	86	98	110	41
LSD (0.05%)	1.72	2.74	4.34	1.84	1.23

TDOP- Total death of plant; DOS- Duration of senescence; *****Statistically significant at P= 0.05.

Weight of grains per pod

The weight of grains per pod of each treatment and control plants were weighed and recorded using a weighing balance. The weighing balance was adjusted to 0. The grains were then removed from the pods and placed on the weighing balance, and the weight the grains recorded.

Weight of grains per plant

The dry weight of the grains of the harvested fruits of all the cowpea varieties giving hormone and fertilizer treatments were recorded as mean weight of seeds per plant and taken as yield per treatment.

Statistical analysis

The data collected was subjected to one-way analysis of variance (ANOVA) using GENSTAT software and means with significant differences were separated using least significance difference test at P<0.05.

RESULTS AND DISCUSSION

Senescence

The result of onset, progression and duration of senescence in the two cowpea varieties, is presented in Table 1. Comparison of the varieties showed significant difference (P≤0.05) in all the parameters. In the variety Kanannado, there was a general delay in the number of days taken to the onset of senescence and days to 50% senescence, days to 90% senescence, and days to total death of the plant, when compared with IT89KD–288 (Table 1).

The effect of the various treatments on senescence is also shown in Table 1. The treatments induced different responses for the number of days taken to the onset of senescence, days to 50% senescence, days to 90% senescence, and days to total death of the plants. Comparison between the treatments and the control showed that nitrogen fertilizer hastened senescence, while BAP treatment applied as a single treatment or in a combination with N fertilizer delayed senescence. Nitrogen fertilizer is not known to delay senescence. Several studies reported that N fertilizer is known to improve soil productivity and fertility which improved yield and quality of crops (Whalen, 2000; Maerere and Ishimine, 2001; Vanek, 2003). Soil treated with N fertilizer was found to be loose and this probably provided adequate aeration in the soil and improved microbial activities (Xio and Li, 2006). On the other hand, turmeric plant when treated with nitrogen fertilizer remained green for a longer time and resulted in a higher vegetative growth and yield (Mazid, 1993; Seobi, 2005; Anes and Johnson, 1980).

Nitrogen treatments (150% N) hastened the onset of senescence by 66 days, days to 50% senescence by 82 days, 90% by 96 days and total death by 105 days. The combined treatment (150% N + 200 ppm BAP) induced

Table 2. Yield (g) and yield attributes of the cowpea varieties, treatments and seasons at first harvest.

Parameters	No of pod per plant	Pod length (cm)	No of grains per pod	Weight of grains per pod(g)	Weight of grains per plant(g)
Variety					
IT89KD-288	10.000^{a}	15.817^{a}	13.000^{a}	3.404^{a}	16.47^{a}
Kanannado	8.000^{b}	14.687^{b}	12.000^{b}	3.175^{a}	18.53^{a}
Mean	9.167	15.252	12.969	3.290	17.50
LSD (0.05%)	0.538	0.370	0.410	NS	NS
Treatments					
150% N	11.000^{a}	17.067^{a}	14.000^{a}	4.042^{a}	23.34^{a}
200 ppm BAP	8.000^{c}	14.525^{c}	12.000^{c}	3.108^{c}	15.58^{c}
150%N + 200 ppm BAP	10.000^{b}	16.472^{b}	14.000^{b}	3.550^{b}	19.21^{b}
Control	6.08^{d}	12.925^{d}	10.000^{d}	2.458^{d}	11.88^{d}
Mean	9.167	15.252	12.969	3.290	17.50
LSD (0.05%)	0.4127	0.448	0.490	0.108	0.934
Season					
Rainy	11.000^{a}	16.179^{a}	13.000^{a}	3.479^{a}	20.35^{a}
Dry	7.000^{b}	14.325^{b}	12.000^{b}	3.100^{b}	14.65^{b}
Mean	9.167	15.252	12.969	3.290	17.50
LSD	0.375	0.375	0.421	0.189	1.227

delay in the onset of senescence by 72 days, 50% senescence by 96 days, 90% senescence by 110 days and total death of plant by 121 days (Table 1). Treatment with 200 ppm BAP also delayed the onset of senescence (77 days), and 50% senescence (97 days), 90% senescence (106 days) and total death of plant (120 days). There was no significant difference between 200 ppm BAP treated plants and the combined treatment of 150% nitrogen fertilizer + 200 ppm BAP with respect to 90% senescence and total death of plants. Several workers such as Nooden (1978) and Richmond and Lang (1994), reported that BAP and gibberellins retard senescence, while abcissic acid and ethylene tend to act as accelerators. Leaf senescence can be retarded locally by the application of BAP (Schuphan, 1974). Physiological studies suggest that BAP can regulate leaf senescence and that the internal BAP level drops with the progression of leaf senescence (Schuphan, 1974). Senescence is the result of complex changes in basic plant metabolism. In higher plants, various degradative phenomena associated with free radicals (FRs) have been implicated in the senescence process (Leshem et al., 1986; Thompson et al., 1987). Onset of senescence in the control plants occurred at 57 days after planting, 50% at 69 days, and 90% at 81 days and total death at 92 days after planting (Table 1). The duration of senescence from its onset to total death of plants ranged from 35 days in the control to 39 days in 150% N to 42 days in 200 ppm BAP to 48 days in 150% N + 200 ppm BAP (Table 1). This shows that reproductive period in the combined treatment of 150% N + 200 ppm BAP was most extended by about 13 days.

Effect of the two planting seasons (rainy and dry) on senescence is presented in Table 1. Onset of senescence, days to 50 and 90% senescence and total death of the plants were earlier for the dry season planting when compared with the rainy season planting with significant difference (P≤ 0.05). In the rainy season, senescence commenced from 69 days after planting (DAP) and total death of the plants occurred by 117 DAP, whereas onset of senescence in the dry season was recorded at 67 DAP and total death at 102 DAP (Table 1). The duration of senescence from onset to total death were 35 days in the dry season and 47 days in the rainy season (Table 1) suggesting that reproductive period was extended by about 12 days in the rainy season.

Yield

There was significant difference between the two varieties of cowpea used in this experiment with respect to yield parameters. In the first harvest, variety IT89KD – 288 had greater number of pods, number of seeds and length of pods. There was exception in weight of grains per pod and weight of grains per plant in which no significant difference was observed (Table 2), but during the second harvest, kanannado variety had greater number of pods, number of seeds, length of pods, weight of grains per pod and weight of grains per plant when compared with IT89KD-288 (Table 3). This might be due to the early commencement of senescence in IT89KD-288 that led to the death of most of the plants before the2nd harvest. However, IT89KD-288 produced less yield

Table 3. Yield (g) and yield attributes of the cowpea varieties, treatments and seasons at second harvest.

Parameters	No of pod per plant	Pod length (cm)	No of grains per pod	Weight of grains per pod (g)	Weight of grains per plant (g)
Variety					
IT89KD-288	4.000^b	11.31^b	9.000^b	2.054^b	11.27^b
Kanannado	5.000^a	12.21^a	10.000^a	2.683^a	12.03^a
Mean	5.229	11.76	9.917	2.569	11.65
LSD (0.05%)	0.179	0.754	0.179	0.471	0.031
Treatments					
150% N	5.000^c	16.29^b	13.000^b	2.892^c	10.20^c
200ppm BAP	7.000^b	14.19^c	11.000^c	3.517^b	14.85^b
150% N + 200 ppm BAP	8.000^a	16.57^a	14.000^a	3.867^a	18.54^a
Control	0.000	0.000	0.000	0.000	0.000
Mean	5.229	11.76	9.917	2.569	11.65
LSD (0.05%)	0.534	0.900	0.722	0.204	0.986
Seasons					
Rainy	5.000^a	12.47^a	10.000^a	2.704^a	11.34^a
Dry	4.000^b	11.05^c	9.000^c	2.433^c	10.96^a
Mean	5.229	11.76	9.917b	2.569d	11.65
LSD	0.153	0.644	0.293	0.142	NS

when compared with kanannado that had a delay in the onset of senescence and thus most of its plants were alive before the 2nd harvest and therefore produced more yield in the 2nd harvest.

Fertilizer and hormonal treatments increased yield in the cowpea varieties grown in the different seasons with significant difference when compared with the control plants. Combined treatment of 150% N fertilizer and 200 ppm BAP treatment induced significantly higher yield in the two harvests (1st and 2nd harvests) as compared to the rest of the treatments. This might be due to the early commencement of senescence in the other treatments that led to the death of the plants before the 2nd harvest which subsequently led to low yield.

The control plants did not have any yield as a result of the death of all the plants before the 2nd harvest (Table 3). It was reported that in cowpea, many plants often die after producing the 1st flush of pods. This causes substantial reduction in total grain yield in that 2nd flush yield is proportional to the number of plants surviving to produce the 2nd flush (Ismail and Hall, 1998.) Several authors have reported the same observations on some cowpea varieties (Ferry and Singh, 1997; Singh, 2002; Boukar et al., 2015). The delayed leaf senescence result from a higher proportion of plants surviving after the production of the 1st flush of pods and probably results from the maintenance of root viability (Gwathmey and Madore, 1992; Fatokun et al., 2013) which could enhance nitrogen fixation.

The single hormone treatments, that is, 200 ppm BAP did not show significant effect on yield when compared with the control plants. Certain hormones have been found to increase yield in many crops, while some other growth substances do not have significant effect on yield. For instance, the auxin, b–naphthoacetic acid (NAA) sprayed on the open flowers of *Solanum melongena* either singly or in combination were observed to increase fruit set as well as total weights of the fruits (Olympios, 1976). Subramanian and Kende (1985) observed that growth substances influenced seed yield in cowpea even though the yield component were not significantly affected.

Kaul et al. (1976) reported that an increase in number of pods/plant by seed treatment with 200 ppm planotix increased yield by 33%. Thomas and Stoddart (1976) reported that auxins have been used for many years to increase fruit set of tomatoes. Several workers showed the importance of N fertilizer, in increasing grain yield and its components. Nour (1998) and El-Kholy et al. (1999) found that application of fertilizer significantly increased grain yield in rice when compared with the control. Ebaid and Ghanem (2000) reported that panicle weight, 1000-grain weight and grain yield in rice were significantly increased as the N fertilizer increased up to 30 tons/ha, while 70 tons/ha was adequate for the highest values of panicle length and number of grain/panicle. Mazid (1993) reported that N fertilizer should be applied in a particular ratio, for higher growth and yield of a specific plant species. El-Batal et al. (2004) showed that increasing N fertilizer rate from 50 to 80 kg N/feed significantly

Table 4. Grain yield at first and second harvest and the total yield.

Treatments	Grain yield at first harvest (g)	Grain yield second harvest (g)	Total yield (g)
150% N	23.34^a	10.20^c	33.57^b
200 ppm BAP	15.58^c	14.85^b	30.43^c
150% N + 200 ppm BAP	19.21^b	18.54^a	37.75^a
Control	11.88^d	0.00	11.88^d
Mean	17.50	14.53	
LSD (0.05%)	0.934	0.986	

increased plant height and yield. Vanek (2003) reported that regular application of N fertilizer to root crops leads to higher yield.

Comparing the 2 seasons during the 1st harvest, greater yield was obtained in rainy season with significant difference (Table 2). In the 2nd harvest, rainy season differed significantly from the dry season in the number of pods, length of pods, number of grains per pod, weight of grains per pod but no significant difference with regards to weight of grains per plant (Table 3).

Wallace (1985) and Summerfield et al. (1975) demonstrated positive relationship between yield and photoperiod temperature response; the increase in yield is due to increase in number of nodes at which pods could be set, indicating that the nodes arose from new branches and continued elongation of existing indeterminate stem and branches. They also reported that plant size at flowering and the number of nodes produced has a great influence on subsequent yield in indeterminate genotypes. They noted that stunted plants due to adverse conditions gave poor yield. Grain yield in cowpea is dependent on both vegetative and reproductive component that are in turn governed by environmental factors such as day length, temperature and soil moisture (Chaudhry and Ogo, 1985). Economic yield is expected to show close positive relation with total plant dry weight, 50% of which to a large extent is dependent on number of leaves, plant height and number and length of branches (Summerfield et al., 1975). As recent examples, Souleymane et al. (2013) and Huynh et al. (2015) confirmed that with the improved variety, IT97K-556-6.

Grain yield at 1st and 2nd harvest and the total yield

Table 4 shows the grain yield at 1st and 2nd harvest and the total yield with respect to the treatments. At 1st harvest, 150% N treated produced greater yield than the combined treatment of 150% N + 200 ppm and then the 200 ppm BAP treatments, and the least were the control plants. In the 2nd yield, 150% N + 200 ppm produced greater yield than the rest of the treatments. Therefore, at the end when the 1st and 2nd harvest were added together, the total yield was greater in 150% N + 200 ppm followed by 150% N then 200 ppm BAP, the least in the total yield were the control plants (Table 4).

Relationship between duration of senescence, 1st and 2nd harvest and the total yield in relation to treatments

The relationship between senescence, 1st and 2nd harvests and the total yield showed that the combined treatments of 150% N + 200 ppm BAP took longer days from the onset of senescence to the total death of the plants followed by the 200 ppm BAP treatment, then 150% N. Therefore, due to the delayed senescence in the 150% N + 200 ppm BAP and single 200 ppm BAP treatments, they were able to produce 2nd flush of pods which led to greater yield in the 2nd harvest and the total grain yield when compared with 150% N treatment that took lesser days from the onset of senescence to the total death of the plants, and therefore produced less yield in the 2nd harvest and total yield, and this shows that the longer the days the plant takes to senesces, the greater the yield (Figure 1). A relationship between crop yield and senescence has been postulated for many years, Thomas et al. (1996) assumed that an extended period of maximal photosynthetic activity, that is, delayed senescence, should lead to higher yields. Whereas a positive correlation between leaf senescence duration and yield may be valid for most crops with regard to total biomass production and for tuber crops; the relationship is more complicated with respect to seed yields. In particular for cereal crops, there has been a long discussion on whether grain yield is determined by sink (the developing grain) or by source (the photosynthesizing vegetative tissues). The predominant current view is that sink strength is the main limiting factor for yields, especially in small-grain cereals such as wheat (Fischer, 2008; Boukar et al., 2013) and, hence, physiological events particularly in the period around seed setting are crucial for determining yield levels. The genetically determined senescence program can indeed have bearings on the productivity of crop plants including seed yields (Borra´s et al., 2004; Huynh et al., 2016).

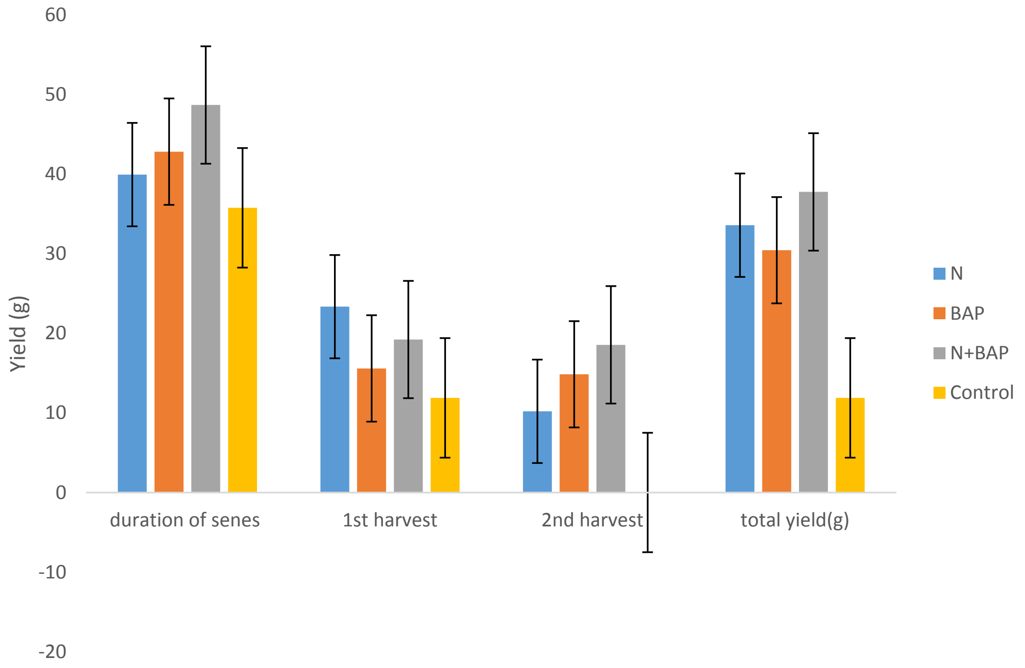

Figure 1. Comparison between duration of senescence, 1st and 2nd harvest and total yield.

The association among senescence parameters and yield are found in sorghum and in particular corn. A classical study on 10 short season maize hybrids showed a positive correlation between leaf senescence duration and grain yield (Tollenaar and Daynard, 1978). Similar relationship between leaf area duration/stay-green and yield parameters were reported for sorghum hybrid lines (Borrell et al., 2000) and also for oilseed rape (Hunkova et al., 2011). In addition to these strong examples, there are many studies involving a number of species which demonstrates that genetic modulation of senescence parameters can indeed affect the yield levels, even though the effects are variable and highly influenced by environmental conditions.

Conclusion

In conclusion, the combined liquid nitrogen fertilizer and BAP regulated the onset of senescence and had higher overall yield when compared with the other treatments for the two cowpea varieties. This therefore implies that the longer the days the plant takes to senesces, the greater the yield. From the findings of this study, the following can be recommended:

1. The use of liquid N fertilizer could be recommended to farmers to improve growth and yield in photosensitive cowpeas.
2. N fertilizer in combination with BAP could be used to extend the reproductive period and the photosynthetic efficiency of the plant, leading to increase in yield as well as delay senescence in the photosensitive cowpea studied.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Full Length Research Paper

Heterosis and combining ability analysis of quality protein maize (*Zea mays* L.) inbred lines adapted to mid-altitude sub-humid agro-ecology of Ethiopia

Dufera Tulu[1*], Bulti Tesso[2] and Girum Azmach[1]

[1]EIAR- Bako National Maize Research Centre, P. O. Box 2003, Addis Ababa, Ethiopia.
[2]Department of Plant Sciences, Haramaya University, Haramaya, Ethiopia.

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Generation of information on heterosis and combining abilities of newly developed maize inbred lines is necessary for a successful hybrid and synthetic maize varieties development. Accordingly, this study was conducted to estimate the combining ability of QPM inbred lines for grain yield and yield related traits and to determine the magnitudes of standard heterosis for grain yield and yield related traits in line × tester QPM hybrids. Fifty test crosses together with two standard checks were evaluated using alpha lattice design with three replications at three mid-altitude sub humid trial sites (Bako, Hawassa and Jimma) in Ethiopia during 2016 main cropping season. Combined analysis of variance showed highly significant differences among the three locations for all the studied traits indicating the presence of considerable variation among locations for genotype performance. The interaction between sites and genotypes were highly significant and significant ($P<0.05$) for grain yield and ear height, indicating that the performances of the genotypes and crosses were not consistent for these traits. The significance of both general combining ability (GCA) and specific combining ability (SCA) mean square for some traits indicates the role of additive and non-additive gene action in the inheritance of the traits. However, for all the traits, the contribution of GCA variance was greater than the contribution of SCA variance, revealing the predominance of additive gene action in the inheritance of all the traits studied. L1 and L3 had significant positive GCA effects and are considered as good combiners for grain yield. In addition, L1 and L9 were good combiners for earliness. In this study, none of the crosses showed positive and significant standard heterosis for grain yield.

Key words: General combining ability, grain yield, specific combining ability, standard heterosis.

INTRODUCTION

Maize is one of the most important field crops cultivated in Ethiopia to ensure food security. Maize contributes the greatest share of production and consumption together with other major cereal crops, such as tef [*Eragrostis tef*

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*Corresponding author. E-mail: duferatulu@gmail.com.

Table 1. Description of the study areas.

Sites	Altitude (masl)	Latitude N	Longitude E	Temperature (°C)		Rainfall (mm)	Soil type
				Min	Max		
Bako	1650	9° 06'	37° 09'	13.3	28.0	1239.4	Nitosol
Hawassa	1689	7° 04'	38° 31'	12.6	27.3	1002.0	Vitric andosol
Jimma	1750	7° 46'	36° 00'	11.2	25.9	1536.0	Nitosol

(Zucc.) Trotter], wheat (*Triticum aestivum* L.) and sorghum [*Sorghum bicolor* (L.) Moench]. Among the cereal crops, maize ranks second in area coverage and first in total annual production and productivity in Ethiopia (CSA, 2016).

Despite its widespread and increased consumption as a source of carbohydrates/energy, maize, like all cereal crops, is known to be poor in its kernel protein quality. The maize protein is limited in two essential amino acids- lysine ($C_6H_{14}N_2O_2$) and tryptophan ($C_{11}H_{12}N_2O_2$) (Bressani, 1991). Protein malnutrition is therefore a serious problem, especially among children, where maize and other cereal crops are the predominant staple foods. Quality protein maize (QPM) is a type of maize variety with improved quality protein content developed after the discovery of maize mutant in the mid 1960's containing the opaque-2 gene which enhances levels of lysine and tryptophan in the endosperm protein (Mertz et al., 1964). Consumption of QPM instead of the conventional maize (CM) that has low protein quality can substantially improve the protein status and greatly reduce the malnutrition problem of impoverished people that are dependent on maize as their staple food (Leta et al., 2003). Cognizant of the potential benefits of QPM varieties, the National Maize Research Program of Ethiopia initiated a systematic QPM research in collaboration with CIMMYT in the early 1990s, which led to the identification and release of the first QPM hybrid, BHQP542 in 2002 (Legesse et al., 2012), Melkassa 6Q in 2008 (Gezahegn et al., 2012) and the subsequent release of other several QPM varieties (Adefris et al., 2015).

Information on combining ability of parental maize inbred lines, that is, general combining ability (GCA) and specific combining ability (SCA), which determine their performances in hybrid combination, is an important input for designing breeding strategy aimed at exploiting the genetic potential of maize for achieving higher productivity (Chawla and Gupta, 1984). Combining ability studies can help understand the type of gene action involved in controlling quantitative characters, thereby assisting breeders in selecting suitable parent materials (Hallauer and Miranda, 1988).

Heterosis is also important in maize breeding and is dependent on level of dominance and differences in gene frequency. The manifestation of heterosis depends on genetic divergence of the two parental varieties (Hallauer and Miranda, 1988). It is manifested as an increase in vigor, size, growth rate, yield or some other characteristics. But in some cases, the hybrid may be inferior to the weaker parent, which is also considered as heterosis. That means heterosis can be positive or negative. The interpretation of heterosis depends on the nature of trait under study and the way it is measured. Generally, heterosis is an important trait used by breeders to evaluate the performance of offspring in relation to their parents. It estimates the enhanced performance of hybrids as compared to their parents. Often, the superiority of F_1 is estimated over the average of the two parents, or the mid parent.

Breeding efforts are underway to convert elite mid-altitude CM inbred lines to QPM through back crossing in recent years in Ethiopia by the breeding program of Bako National Maize Research Center (BNMRC) of the Ethiopian Institute of Agricultural Research (EIAR). This effort has led to the development of many QPM inbred lines, including inbred lines used in this study. Thus, this study was conducted to estimate the combining ability of QPM inbred lines for grain yield and yield related traits and magnitudes of standard heterosis for grain yield and yield related traits in line x tester QPM hybrids.

MATERIALS AND METHODS

Description of experimental sites

The study was conducted at three locations in the mid-altitude sub-humid agro ecologies of Ethiopia, namely, Bako, Hawassa and Jimma Agricultural Research Centers in the main cropping season of 2016 (Table 1).

Experimental materials

A total of 52 entries composed of 50 test crosses, formed by crossing 25 QPM inbred lines with two single cross testers (referred to as tester A and tester B), and two standard checks (BHQPY545, yellow QPM and BH546, white CM) were studied. The QPM inbred lines were previously developed by BNMRC through backcross breeding technique using elite CM inbred lines as recurrent parents and elite QPM lines as donor parents. The list and the pedigrees of the inbred lines used in the line by tester crosses and that of the testers are given in Table 2. A standard QPM conversion procedure developed by CIMMYT was used to develop the QPM inbred lines, which involved kernel light table screening for endosperm modification, laboratory analysis for tryptophan and lysine contents, as well as field evaluation for agronomic traits. The testers used in this study were identified by CIMMYT Zimbabwe and introduced to Ethiopia by BNMRC breeding program in 2014 main season.

Table 2. List of QPM inbred lines selected and used for cross formation and testers.

S/N	Lines code	Pedigree	Origin (source)
1	L1	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-12-1-2-1-1-1	BNMRC
2	L2	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-20-1-1-1-1-1	>>
3	L3	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-12-1-2-2-1-1	>>
4	L4	(CML-144 X SC-22(F2) x SC-22(F2) x SC-22)-B-44-2-1-2-1-1	>>
5	L5	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-12-1-1-2-1-1	>>
6	L6	(CML-144 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-25-1-1-1-1-2	>>
7	L7	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-40-1-1-1-1-1	>>
8	L8	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-15-1-2-2-1-1	>>
9	L9	(CML-144 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-32-1-1-2-1-3	>>
10	L10	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-12-1-3-3-1-1	>>
11	L11	(CML-144 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-25-1-1-1-1-1	>>
12	L12	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-12-1-3-2-1-1	>>
13	L13	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-12-1-3-1-2-1	>>
14	L14	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-20-1-1-3-1-1	>>
15	L15	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-15-1-1-1-1-1	>>
16	L16	BK02-Z-311-28(F2)-B-1 X CML-144(F2)-15-2-3-1-1	>>
17	L17	BK02-Z-311-28(F2)-B-1 X CML-144(F2)-15-1-1-1-1	>>
18	L18	BK02-Z-311-28(F2)-B-1 X CML-144(F2)-48-1-1-1-1	>>
19	L19	BK02-Z-311-28(F2)-B-1 X CML-144(F2)-15-2-1-2-1	>>
20	L20	BK02-Z-311-28(F2)-B-1 X CML-144(F2)-15-2-3-2-1	>>
21	L21	(CML-144 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-32-1-1-2-1-1	>>
22	L22	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-15-1-2-1-1-1	>>
23	L23	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-15-1-1-2-1-1	BNMRC
24	L24	(CML-142 X 144-7-b(F2) x 144-7-b(F2) x 144-7-b)-B-15-1-2-3-1-1	>>
25	L25	(CML-144 X SC-22(F2) x SC-22(F2) x SC-22)-B-44-2-1-1-1-1	>>
26	T1	CML144/CZLQ5	CIMMYT
27	T2	CZLQ2/CML511	>>

*BNMRC = Bako National Maize Research Center.

Experimental design and field managements

The experimental design was (0, 1) alpha lattice design (Patterson and Williams, 1976) with 4 plots per an incomplete block and 13 incomplete blocks with three replicates. Each entry was planted in a two row 5.1 m long plot with spacing of 0.75 m between rows and 0.30 m between plants within a row. The experimental materials were hand planted with two seeds per hill, which were later thinned to one plant to get the recommended planting density for the testing sites, 44,444 plants per hectare. Planting was conducted on the onset of the main rainy season after an adequate soil moisture level was reached to ensure good germination and seedling development. Other agronomic practices were carried out as per the recommendation for the test areas.

Data collection

Data on grain yield and other important agronomic traits were collected on a plot and sampled plants/ears bases. Data collected on a plot basis include days to 50% anthesis, days to 50% silking, number of ears per plant, actual moisture content, field weight (kg/plot), plant aspects, ear rot and bad husk cover; while data recorded on sampled plants basis were ear height (cm) and plant height (cm). Yield in t/ha was calculated using CIMMYT fieldbook software (Banziger and Vivek, 2007).

Data analysis

Analysis of variance

Data were subjected to analyses of variance (ANOVA) using the PROC MIXED procedure in SAS$^{®}$ computer program (SAS Institute, 2004). Entries were used as fixed factor while replications and incomplete blocks within replication were considered as random factors. Least significant difference (LSD) was used for mean separation. For traits that displayed significant differences among crosses, line by tester analysis was performed to further partition the variances due to crosses into lines, tester and line by tester effects (Dabholkar, 1999; Singh and Chaudhary, 1985) using SAS program (SAS institute, 2004).

Line by tester analysis

Line by tester analyses was performed for traits that showed significant differences among genotypes as suggested by Dabholkar (1999) and Singh and Chaudhary (1985) to partition the mean square due to crosses into lines (denoting GCA due to lines or males, GCAm), tester (denoting GCA due to testers or females, GCAf) and line x tester interactions (denoting SCA of lines by testers crosses, SCAmf). The following mathematical model was used for the combining ability analysis:

Table 3. Combined analysis of variance for the tested traits in a line by tester mating between 25 QPM maize inbred lines and two testers evaluated at three locations in Ethiopia in 2016.

Source of var.	DF	GY	DA	DS	PH	EH	PA	ER	HC	EPP
Site	2	166.64**	290.9**	716.84**	180918.78**	48294.64**	14.61**	277.34**	14.42**	5.59**
Rep(Site)	4	47.91**	17.69**	34.55**	3582.19**	3111.41**	0.14	0.73*	0.14	0.22**
Block(Rep)	36	1.89	3.13**	3.89**	317.49	222.8	0.15	1.24*	0.79	0.04
Genotypes (G)	51	7.69**	9.77**	13.45**	588.74**	384.69**	0.17*	1.24**	4.32**	0.20**
Crosses (Cr)	49	8.22**	10.26**	14.65**	657.47**	395.41**	0.18*	1.03	4.68**	0.13**
GCA(lines)	24	8.49**	8.84**	14.51**	1078.65**	661.91**	0.19	1.20	7.83**	0.22**
GCA(testers)	1	0.26	157.24**	179.24**	3099.47**	696.89*	0.002	0.22	0.91	0.07
SCA (L*T)	24	8.29**	5.56**	7.93**	134.53	116.35	0.18	0.88	1.67*	0.04
Site*Genotypes	102	5.39**	1.96	2.87	303.41	242.69*	0.14	0.90	1.07	0.04
Site*Crosses	98	6.16**	2.39	3.22*	337.27*	282.07**	0.15	0.76	1.24**	0.04**
Site*GCA(lines)	48	7.46**	2.78*	3.34	427.48**	316.74**	0.17	0.88	1.64**	0.04
Site*GCA(testers)	2	9.04**	7.20*	8.10*	281.25	668.90*	0.03	1.49	1.85	0.06
Site* SCA(L*T)	48	4.73**	1.81	2.91	249.39	231.28	0.12	0.62	0.81	0.05*
Pooled error G.	270	3.14	1.70	2.23	242.32	171.76	0.12	0.75	0.98	0.05
pooled error Cr.	294	2.80	1.92	2.45	252.75	176.97	0.13	0.82	0.98	0.03
CV (%)		22.25	1.59	1.79	6.18	9.35	13.27	30.30	31.67	17.56
Cont. of GCA		50.62	73.48	73.48	89.98	85.59	51.25	58.53	80.64	84.66
Cont. of SCA		49.38	26.52	26.52	10.02	14.41	48.75	41.47	19.36	15.34

*=0.05 and **= 0.01 significant probability level respectively.GY=grain yield, DA = days to anthesis, DS = days to silking, EH = ear height, PH = plant height, PA=plant aspect, ER=ear rot, HC=husk cover, EPP = number of ears per plant, GCA = general combining ability; SCA = specific combining ability; DF = degrees of freedom, Cont. of GCA = contribution of general combining ability of lines and testers, Cont. of SCA = contribution of specific combining ability of line by tester.

$$Y_{ijk} = \mu + r_k + g_i + g_j + S_{ij} + e_{ijk}$$

Where, Y_{ijk} = the value of a character measured on cross of line i by tester j in kth replication; μ = population mean; r_k= effect of kth replication; g_i = general combining ability (GCA) effects of ith line; g_j = general combining ability (GCA) effect of the jth tester; S_{ij} = specific combining ability (SCA) of ith line and jth testers such that S_{ij} equals S_{ji}; e_{ijk} = experimental error for ijkth observation.

GCA and SCA of lines were computed for characters that showed significant differences among crosses following line by tester (LxT) analysis as suggested by Singh and Chaudhary (1985). The proportional contributions of lines (GCA_L), testers (GCA_T), and their interaction (SCA_{LxT}) with the sum square of crosses were calculated as the ratio between sum of squares of each component and the cross sum of squares as given by Singh and Chaudary (1985) as follows:

Contribution of lines (L) = $\frac{\text{SS(l)}}{\text{SS(Crosses)}} \times 100$

Contribution of testers (T) = $\frac{\text{SS(t)}}{\text{SS(Crosses)}} \times 100$

Contribution of line by tester (L x T) = $\frac{\text{SS(lxt)}}{\text{SS(Crosses)}} \times 100$

The significance of GCA and SCA effects were tested by dividing the corresponding SCA and GCA values by their respective standard error, to obtain the calculated t values, and comparing the calculated t value with tabular t-value at the error degree of freedom**.**

Standard heterosis (SH) in percent was calculated for those traits that showed statistically significant differences among genotypes as suggested by Falconer and Mackay (1996). These were computed as percentage increase or decrease of the cross performances over best standard check as follows:

$$SH(\%) = \frac{F1 - SV}{SV} * 100$$

Where, F1 = mean value of a cross; SV = mean value of standard check variety.

Test of significance for heterosis was done using the t-test. The standard errors of the difference for heterosis were calculated as follows:

SE(d) for SH = $\pm\sqrt{2\text{MSE}/\text{r}}$

Where, *SE (d)* is standard error of the difference, MSe is error mean square and r is number of replications and calculated t value was compared against the tabulated t-value at degree of freedom for error.

t (standard check) = F1– SV/SE(d)

RESULTS AND DISCUSSION

Analysis of variance

Combined analysis of variance showed highly significant differences among the three locations for all the studied traits (Table 3). The result also showed highly significant (P<0.01) mean squares due to genotypes for all characters studied.

Table 4. Mean grain yield and agronomic traits of top-yielding QPM hybrids and standard checks evaluated across three locations of mid-altitude agro-ecologies in Ethiopia.

Crosses	Grain yield (t/ha)				DA (days)	DS (days)	PH (cm)	EH (cm)	PA (1-5)	ER (%)	HC (%)	EPP (#)
	Bako	Hawassa	Jimma	Across								
L1xT1	12.09	11.22	9.29	10.25	83.22	85.11	263.33	143.33	2.44	1.91	3.51	1.36
L23xT1	9.95	11.52	9.86	9.81	82.33	83.89	254.78	140.33	2.78	2.91	3.82	1.31
L3xT1	12.69	9.83	8.32	9.80	84.00	86.00	260.11	145.33	2.56	2.23	3.62	1.30
L9xT2	11.62	9.68	9.44	9.75	82.56	85.00	242.11	131.56	2.72	2.33	1.18	1.23
L22xT2	9.78	8.18	10.16	9.57	79.44	80.78	258.56	146.56	2.50	1.61	3.58	1.06
L20xT2	9.26	6.91	10.98	9.14	80.00	81.11	243.22	131.33	2.39	1.98	1.40	1.51
L15xT1	11.15	10.36	6.91	9.09	82.11	84.44	272.22	152.00	2.67	2.48	3.56	1.29
L13xT2	9.92	7.75	9.26	9.03	80.22	81.89	256.33	144.00	2.56	2.24	1.33	1.39
L3xT2	10.70	8.06	7.55	8.80	81.22	82.78	252.11	142.44	2.56	3.27	3.00	1.28
L11xT1	10.52	8.58	7.76	8.79	82.00	83.78	261.11	152.22	2.56	2.97	2.49	1.21
BHQPY545	10.21	8.57	8.72	9.33	80.33	81.67	240.56	127.11	2.50	3.24	1.04	2.05
BH546	10.50	8.27	7.44	8.49	80.00	81.89	253.56	132.22	2.89	2.43	1.72	1.22
Mean	8.55	8.31	7.95	7.97	81.82	83.44	251.94	140.19	2.64	2.28	2.43	1.28
LSD	1.64	2.35	3.38	1.64	1.21	1.38	14.45	12.16	0.32	1.04	1.15	0.21
Max	12.69	11.52	11.70	10.25	84.00	86.11	272.22	153.22	3.06	3.27	4.10	2.05
Min	5.27	5.53	6.28	5.97	79.44	80.78	231.56	124.33	2.33	1.51	0.91	1.06

DA = Days to anthesis, DS = days to silking, EH = ear height, PH = plant height, PA=plant aspect, ER=ear rot, HC=husk cover, EPP = number of ears per plant, LSD = least significant difference.

The interaction between sites and genotypes (S x G), were highly significant and significant ($P< 0.05$) for grain yield, days maturity and ear height, indicating that the performances of the genotypes and crosses were not consistent for these traits. However non significant interaction effects of S x G were observed for most of the traits, indicating that the genotypes were performed uniformly across sites for those traits. Generally, the traits which showed significant S x G interaction had a differential genotypic response to variable environmental conditions and this resulted in change in the ranks of genotypes and limited the identification of superior genotypes for all sites. This revealed the site specificity of the genotypes tested (Bayisa et al., 2008).

Mean performance of genotypes

The mean performances of the genotypes (the 50 hybrid progenies and two checks) across site are given in Table 4. The mean grain yields (GY) of the genotypes across sites ranged from 5.97 to 10.25 t/ha with overall mean of 7.97 t/ha. The L1 x T1, which was the highest yielding cross (10.25 t/ha) out yielded the high yielding check, BHQPY545 (9.33 t/ha) by 9.86% and the other check BH546 (8.49) by 20.73%. The presence of crosses having mean values better than the standard checks indicate the possibility of obtaining good hybrid (s) for future use in breeding program or for commercial use.

Days to anthesis and silking ranged from 79.44 to 84.00 and 80.78 to 86.11 days, with overall means of 81.82, and 83.44 days, respectively. The shortest numbers of days were recorded for crosses L22 x T2 (79.44) days to anthesis and L22 x T2 (80.78) days to silking (Table 3). Most of the crosses showed longest number of days to anthesis and silking. This shows that those crosses could be grouped as late maturing types. Late maturing crosses are important in the breeding programs for development of high yielding hybrids in areas that receive sufficient rain fall (Girma et al., 2015). Further evaluation and recommendation of this group of materials should be based on agro-ecological suitability.

Plant and ear height ranged from 231.56 to 272.22 and 124.33 to 153.22 cm with mean values of 251.94 and 140.19 cm, respectively. The lowest mean values for both plant and ear heights were observed for the cross L19 x T2, while the highest mean values were measured from the crosses L15 x T1 for plant height and L6 x T1 for ear height. Two crosses were significantly taller than the check BH546. Of these crosses, L15 x T1 gave higher grain yield than the best check BH546 (253.56 cm). In line with this finding, Girma et al. (2015) reported higher GY from taller plants and the authors also suggested that this could be attributed to high photosynthetic products accumulation during long period for grain filling.

Number of ears per plant ranged from 1.06 (L22 xT2) to 2.05 (BHQPY545), with an overall mean of 1.28. Among the top yielding crosses, only L20 x T2 had the number of ears per plant greater than 1.5. The mean performances of hybrids for plant aspect (PA) ranged from 2.33 to 3.06, with an overall mean of 2.64. The high yielding cross L1 x T1 was scored 2.44, While the worst (unattractive) PA scored 3.06 was observed from L6 x T2.

The mean percentages of ear rot (ER) damage among the hybrids ranged from 1.51 to 3.27%. In general, all the crosses and both standard checks showed small percentage score for ER, which means they could be taken as resistant to this disease under natural infestation. Regarding bad husk cover (HC), the percentage mean value range from 0.91 to 4.1%, with overall mean of 1.15%. Nearly all the crosses evaluated in these trials were free of bad husk cover problem.

Combining ability

In the combined analysis of variance, mean squares due to lines (GCAm) were highly significant for all the studied traits, except plant aspect and ear rot, while mean squares due to testers (GCAf) were highly significant for days to anthesis, days to silking, plant height and ear height. Furthermore, mean squares due to lines by tester interaction (SCAmf) of crosses were significant for grain yield, days to anthesis, days to silking and bad husk cover. The significance of both GCA and SCA mean squares for some traits indicates the role of additive and non-additive gene action in the inheritance of these traits (Table 3). Therefore, recurrent selection which exploits both additive and non-additive gene effects simultaneously could be useful in genetic improvement of the traits studied. However, for all of the traits, the contribution of GCA variance was greater than the contribution of SCA variance, revealing the predominance of additive gene action in the inheritance of all traits. This showed that parents with good GCA and *per se* performance could be used to predict the performance of their crosses. Therefore, these parents can be crossed to develop high-yielding QPM hybrids that can potentially be used in further breeding work (inbred line development) and/or directly released for commercial use. Similar results were reported by other authors in their study on combining ability for yield and yield related traits in maize (Bayisa et al., 2008; Chandel and Mankotia, 2014; Seyoum et al., 2016).

Highly significant and significant variations were observed due to interaction between sites and GCA of lines and testers for grain yield, days to anthesis and ear height, indicating that the GCA of inbred lines and testers were affected by the environmental conditions under which the hybrids were grown. SCA x site interaction mean square was only highly significant for the grain yield and significant ($P< 0.05$) for ears per plant.

General combining ability effects

Estimates of GCA effects due to lines and testers various traits combined over site are presented in Table 5. Out of the 25 inbred lines studied in line x tester cross, only two exhibited positive and significant GCA effects for grain yield, while one inbred line displayed negative and significant GCA effects for the same trait (Table 5). L1 and L3 had significant positive GCA effects and are considered as desirable good combiner; while only L18 had significant negative GCA effects and considered as undesirable/poor combiner. However, high positive, non-significant and desirable GCA effects were also revealed by L9 and L13. The significant positive GCA effect of lines indicates the potential advantage of the parents for developing high-yielding hybrids. Similar results were reported by various researchers (Kanagarasu et al., 2010; Beyene et al., 2011; Girma et al., 2015; Ram et al., 2015). For days to anthesis, L1 and L9, and days to silking, L1, L9 and L21 showed positive and significant GCA effects, while negative and significant GCA effects were observed for L22 for days to anthesis and for L20 and L22 for days to silking. Lines with negative GCA effects for days to anthesis and days to silking are desirable lines, as these lines tend to flower earlier than other lines. Even though there is adequate rainfall in mid altitude agro-ecologies of Ethiopia, effort should be made to develop early maturing varieties to fit fluctuating weather condition. Thus, there is possibility of making effective selection for these traits, which could lead to considerable genetic improvement for earliness. Desirability of negative GCA for days to anthesis and silking was suggested by various authors' (Iqbal et al., 2007; Shushay et al., 2013; Umar et al., 2014). In addition, T1 had positive and highly significant GCA effects on both days to anthesis and silking, while T2 revealed negative and highly significant GCA effects for both days to anthesis and silking, indicating T2 is a desirable tester for making earliness when crossed with other lines.

The GCA estimates of lines ranged from -2.88 to 1.73 for maturity date (DM). Only L20 showed negative and significant GCA effects for this trait. Inbred lines that showed negative GCA effects for DM could be considered as good general combiners for developing early maturing hybrids to escape late coming disease and pest infestation as well as terminal moisture stress. In line with the current study, other authors reported both positive and negative GCA effects of inbred lines for DM (Habtamu, 2015; Ram et al., 2015). In addition, Girma et al. (2015) reported significant negative and positive GCA effects for DM and suggested that lines with highly significant GCA effects in the negative direction could be used in breeding programs for the introgression of gene for early maturation.

For PH and EH, L16, L17, L18, L19 and L20 showed negative GCA effects. For EH, positive significant GCA estimates was observed for L6, while L15, showed positive and significant GCA for PH. Negative GCA effects for EH and PH indicates shorter plant height and lower placement of ear, which is very important for development of genotypes resistant to lodging. Therefore, inbred lines with significant negative GCA

Table 5. Estimates of general combining ability (GCA) effects for grain yield and other agronomic traits of 25 maize inbred lines crossed using line x tester mating design and evaluated across site in 2016 main cropping season.

Lines	Characters						
	GY	DA	DS	PH	EH	HC	EPP
L1	1.42*	1.22*	1.43*	8.09	-1.45	0.11	0.09
L2	-0.56	-0.89	-1.01	-4.25	-0.61	0.87*	-0.11
L3	1.37*	0.72	0.88	3.98	3.28	0.84*	0.03
L4	0.24	0.5	0.54	-1.25	-3.50	0.33	-0.05
L5	0.11	0.5	0.21	0.09	-5.39	0.79	0.03
L6	-0.7	0.22	0.82	4.53	11.11*	-0.75	-0.17*
L7	-0.39	0.89	1.10	-1.69	-0.78	-0.51	0.01
L8	-0.51	-0.78	-0.62	4.75	1.39	0.49	-0.06
L9	0.77	1.28*	1.6*	-7.3	-4.72	-1.43**	-0.08
L10	-0.22	0.11	-0.12	0.09	-2.06	-0.25	0.00
L11	0.2	-0.17	-0.01	4.48	7.33	0.10	-0.01
L12	0.23	-0.11	-0.07	1.36	0.61	-0.34	-0.02
L13	0.71	-0.89	-0.9	4.36	2.66	-0.52	0.09
L14	-0.35	-0.06	-0.01	-5.52	3.00	0.52	-0.03
L15	0.54	-0.17	0.38	15.53*	8.55	1.25**	-0.03
L16	-0.17	-0.33	-0.84	-9.25	-5.89	-1.17**	0.32**
L17	-0.65	-0.17	-0.73	-11.69	-10.72*	-0.09	0.17*
L18	-1.38*	0.61	0.82	-6.19	-4.89	-0.37	-0.02
L19	-0.63	-0.11	-0.96	-16.25*	-13.45*	-0.18	0.12
L20	0.02	-0.94	-1.51*	-11.8	-6.56	-0.96*	0.14*
L21	-0.73	1.00	1.38*	-2.25	1.5	-1.45**	-0.04
L22	0.43	-1.28*	-1.29*	10.25	9.16	0.37	-0.15*
L23	0.68	-0.28	0.04	3.25	2.33	1.49**	-0.02
L24	0.25	-0.5	-0.46	9.31	5.44	0.50	-0.15*
L25	-0.7	-0.39	-0.68	7.36	3.66	0.36	-0.06
SE(±)	0.68	0.57	0.64	6.49	5.43	0.40	0.07
SE(gi-gj)	0.97	0.80	0.90	9.18	7.68	0.57	0.10
Testers GCA							
T1	0.02	0.59**	0.63**	2.62	1.24	-0.04	0.01
T2	-0.02	-0.59**	-0.63**	-2.62	-1.24	0.04	-0.01
SE(±)	0.19	0.16	0.18	1.84	1.54	0.11	0.02
SE(gi-gj)	0.27	0.23	0.26	2.60	2.17	0.16	0.03

*=0.05 and **= 0.01 significant probability level respectively. GY=grain yield, DA = days to anthesis, DS = days to silking, EH = ear height, PH = plant height, HC=husk cover, EPP = number of ears per plant, SE = standard error of general combining ability effects of lines and testers, SE (gi-gj)=standard error of the difference of general combining ability effects of lines and testers.

effects are good combiners for hybrid development. Similar results were reported by several authors (Bhatnagar et al., 2004; Aminu and Izge, 2013; Alamerew and Warsi, 2015; Seyoum et al., 2016). About half of the inbred lines revealed positive GCA effects for ear aspect. However, none of the parents showed significant GCA effects for this trait and found to be a good general combiner for quality ear. On the other hand, only L22 revealed significant negative GCA effect for EA. Therefore, L22 is a poorest general combiner for this trait relatively.

In case of bad husk cover (HC), L2, L3, L15, and L23 showed positive and significant GCA effects, while negative and significant GCA effects were observed for L9, L16, L20 and L21. A significant negative GCA effect for HC indicates having closed (firm) husk cover and considered as a good combiner in the desired direction (Girma et al., 2015). Regarding ears per plant (EPP), L16, L17, and L20 revealed significant positive GCA effects for EPP, whereas L6, L22 and L24 showed significant negative GCA effects for the same trait. The positive and significant GCA effect for number of ears per plant indicates prolificacy, which is a desirable trait in increasing maize productivity to some extent (Aminu and Izge, 2013; Alamerew and Warsi, 2015).

Specific combining ability effects

Specific combining ability effects computed for grain yield and other agronomic traits are presented in Table 6. Crosses evaluated in the current study showed limited variation in SCA effects for the traits studied.

For GY, crosses L9 x T2, L16 x T2, L20 x T2, L22 x T2 and L23 x T1 revealed highest positive but non-significant SCA effects with SCA values of 1.08, 0.95, 1.21, 1.24, and 1.18, respectively. This indicates that inbred lines involved in these crosses are genetically divergent, and hence could be regarded to be from different heterotic groups. L9 x T1, L16 x T1, L20 x T1, L22 x T1 and L23 x T2 showed lowest negative but non-significant SCA effects for this trait, indicating that these crosses were poor specific combiners for grain yield. All crosses that showed the highest positive SCA effects, except L23 x T1, resulted from poor inbred lines by poor tester for grain yield. 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. Non significant SCA effect for grain yield was previously reported by Seyoum et al. (2016). In contrast to this finding, Bullo and Dagne (2016) reported highly significant positive and negative SCA effects for GY and they suggested that, when high yielding specific combinations are desired, especially in hybrid maize development, SCA effects could help in the selection of parental material for maximum exploitation of heterosis.

For days to anthesis and silking, only a few crosses showed significant SCA effects in both directions. L5 x T2 showed significant and positive SCA estimate, while L5 x T1 showed significant SCA estimate for DA for both traits. The hybrids with low SCA for days to anthesis and days to silking are desirable as they have earlier anthesis and silking days than what is expected based on GCA of their parents. This finding is in agreement with Kanagarasu et al. (2010), Dagne et al. (2011), Aminu and Izge (2013) and Aminu et al. (2014).

None of the inbred lines had significant SCA effect in both directions for bad husk cover. The result demonstrated that most of the crosses evaluated in the current study did not significantly deviate from what would have been predicted based on their parental performance for almost all the traits. This is expected since the proportions of GCA effects were higher than that of SCA.

Standard heterosis

The estimates of standard heterosis over the standard checks were computed for grain yield and yield related traits and presented in Table 6. None of the crosses showed positive and significant heterosis over both standard checks for grain yield. Standard heterosis (SH) for this trait ranged from -29.62 (L18 x T1) to 20.71% (L1 x T1) over BH546, and -35.97 (L18 xT1) to 9.81% (L1 x T1) over BHQPY545. L3 x T1 (15.46 and 5.04%) and L23 x T1 (15.61 and 5.18%) also exhibited positive standard heterosis over both checks. Positive heterosis for this trait indicates increased yield advantage over the existing standard check. Maize hybrids that perform better than the checks could be used for release as hybrid variety after verification.

Standard heterosis for DA ranged from -0.69 to 5.00% over BH546 and -1.11 to 4.56% over BHQPY545. For DS, the value of SH ranged from -1.36 to 5.16% over BH546 and -1.09 to 5.44% over BHQPY545. None of the crosses displayed negative and significant SH for DA and DS over both checks. On the other hand, 14 crosses showed positive and significant heterosis for DA and DS over both checks indicating, those crosses were late maturing as compared to the checks. Heterosis in the negative direction for these traits indicates earliness of the crosses over the standard checks. In contrast to the current finding, Amiruzzaman et al. (2013) and Bello and Olawuyi (2015) reported negative and significant SH for DA and DS in most of the crosses. Standard heterosis for bad husk cover (HC) ranged from -47.32 to 138.22% and -12.82 to 294.23% for BH546 and BHQPY545, respectively. The negative heterosis in this trait indicates desirable crosses with closed ear up to the tip of the cob, while crosses with positive heterosis for this trait showed bad husk cover and may be susceptible to ear rot disease and are predisposed to other damages.

Conclusion

Combined analysis of variance indicated the presence of considerable variation among locations for genotypes performances. Furthermore, mean squares due to GCA of lines and SCA of crosses were significant for grain yield, days to anthesis, days to silking and bad husk cover. The significance of both GCA and SCA mean squares for these traits, indicate the role of additive and non-additive gene action in the inheritance of these traits. L1, L3, L9 and L13 were identified as good combiner for grain yield and L22 and T2 were identified as good combiner for reducing days to anthesis and silking. The inbred lines having significant negative GCA for days to anthesis and silking identified in this study could be used as parents for breeding quality protein maize for earliness in the mid-altitude sub- humid agro-ecology of Ethiopia. Likewise, L9 x T2, L16 x T2, L20 x T2, L22 x T2 and L23 x T1 revealed highest positive SCA effects.

This indicates that inbred lines involved in these hybrids are genetically divergent, and hence could be regarded to be from different heterotic groups. though, none of the crosses showed positive and significant

Table 6. Estimates of specific combining ability (SCA) effects and standard heterosis (SH) for grain yield and other agronomic traits of 25 maize inbred lines crossed in line x tester mating design and evaluated across sites in 2016 main cropping season.

Hybrids	Grain yield (t/hec)			Days to anthesis (days)			Days to silking (days)			Bad husk cover (%)		
		SH			SH			SH			SH	
	SCA	BH546	BHQPY545	SCA	BH546	BHQPY545	SCA	BH546	BHQPY545	SCA	BH546	BHQPY545
L1xT1	0.87	20.71	9.81	-0.48	4.03**	3.6**	-0.46	3.93**	4.22**	0.97	103.94*	237.5**
L1xT2	-0.87	-0.43	-9.42	0.48	3.75**	3.32*	0.46	3.53*	3.81*	-0.97	-4.58	57.9
L2xT1	-0.11	-14.22	-21.96	0.41	2.50	2.07	-0.02	1.49	1.77	0.39	114.14*	254.38**
L2xT2	0.11	-12.12	-20.05	-0.41	0.00	-0.41	0.02	0.00	0.27	-0.39	73.79	187.61*
L3xT1	0.48	15.46	5.04	0.80	5.00**	4.56**	0.98	5.02**	5.31**	0.35	110.2*	247.87**
L3xT2	-0.48	3.65	-5.70	-0.80	1.53	1.11	-0.98	1.09	1.36	-0.35	74.12	188.14*
L4xT1	-0.24	-6.32	-14.78	0.35	4.17**	3.73**	0.2	3.66*	3.95**	-0.17	50.81	149.58
L4xT2	0.24	-1.33	-10.24	-0.35	1.81	1.38	-0.2	1.63	1.90	0.17	74.56	188.88*
L5xT1	0.59	1.96	-7.24	-1.76*	1.53	1.11	-1.91*	0.68	0.95	0.39	109.56*	246.8**
L5xT2	-0.59	-12.57	-20.46	1.76*	4.44**	4.01**	1.91*	3.80*	4.08**	-0.39	68.56	178.95*
L6xT1	0.88	-4.21	-12.86	-0.37	2.92*	2.49	-0.41	3.26*	3.54*	-0.21	-14.59	41.35
L6xT2	-0.88	-25.41	-32.14*	0.37	2.36	1.94	0.41	2.71	2.99*	0.21	14.27	89.11
L7xT1	0.08	-9.99	-18.11	0.63	5.00**	4.56**	0.65	4.88**	5.17**	-0.60	-23.3	26.92
L7xT2	-0.08	-12.38	-20.29	-0.63	1.94	1.52	-0.65	1.76	2.04	0.60	50.68	149.36
L8xT1	0.37	-7.98	-16.29	-0.15	1.94	1.52	-0.08	1.90	2.18	0.20	80.89	199.36*
L8xT2	-0.37	-17.25	-24.72	0.15	0.83	0.42	0.08	0.54	0.82	-0.20	62.62	169.13*
L9xT1	-1.08	-10.07	-18.18	0.02	4.72**	4.29**	-0.52	4.07**	4.35**	-0.10	-47.32	-12.82
L9xT2	1.08	14.89	4.52	-0.02	3.19*	2.77*	0.52	3.8*	4.08**	0.10	-31.63	13.14
L10xT1	-0.04	-9.46	-17.63	0.41	3.75**	3.32*	0.54	3.26*	3.54*	0.19	37.57	127.67
L10xT2	0.04	-8.98	-17.19	-0.41	1.25	0.83	-0.54	0.41	0.68	-0.19	20.08	98.72
L11xT1	0.64	3.56	-5.79	-0.31	2.50	2.07	-0.35	2.31	2.59	-0.04	44.93	139.85
L11xT2	-0.64	-11.98	-19.92	0.31	1.81	1.38	0.35	1.63	1.90	0.04	53.65	154.27*
L12xT1	-0.18	-5.81	-14.31	-0.04	2.92*	2.49	0.15	2.85	3.13*	-0.65	-16.14	38.78
L12xT2	0.18	-2.07	-10.91	0.04	1.53	1.11	-0.15	0.95	1.22	0.65	63.52	170.62*
L13xT1	-0.42	-2.84	-11.61	0.19	2.22	1.8	0.09	1.76	2.04	0.66	49.00	146.58
L13xT2	0.42	6.4	-3.2	-0.19	0.28	-0.14	-0.09	0.00	0.27	-0.66	-22.85	27.67
L14xT1	0.41	-5.63	-14.15	0.13	3.19*	2.77*	0.42	3.26*	3.54*	0.06	75.02	189.63*
L14xT2	-0.41	-15.8	-23.4	-0.13	1.39	0.97	-0.42	0.68	0.95	-0.06	72.05	184.72*
L15xT1	0.59	7.07	-2.6	-0.2	2.64*	2.21	-0.08	3.12*	3.40*	-0.12	106.65*	241.99**
L15xT2	-0.59	-7.5	-15.85	0.2	1.67	1.24	0.08	1.76	2.04	0.12	124.98**	272.33**
L16xT1	-0.95	-19.5	-26.77	-0.26	2.36	1.94	0.04	1.76	2.04	0.05	-23.89	25.96
L16xT2	0.95	2.21	-7.01	0.26	1.53	1.11	-0.04	0.14	0.41	-0.05	-25.37	23.50
L17xT1	-0.63	-21.43	-28.52	0.46	3.47**	3.04*	0.7	2.71	2.99*	-0.36	15.11	90.49

Table 6. Contd.

L17xT2	**0.63**	**-7.17**	**-15.55**	**-0.46**	**0.83**	**0.42**	**-0.7**	**-0.54**	**-0.27**	**0.36**	**61.46**	**167.2***
L18xT1	-0.60	-29.62	-35.97*	0.8	4.86**	4.43**	1.15	5.16**	5.44**	0.11	26.02	108.55
L18xT2	0.60	-16.02	-23.60	-0.8	1.39	0.97	-1.15	0.81	1.09	-0.11	17.3	94.13
L19xT1	-0.16	-15.68	-23.29	-0.15	2.78*	2.35	0.26	1.9	2.18	-0.30	13.04	87.08
L19xT2	0.16	-12.45	-20.35	0.15	1.67	1.24	-0.26	-0.27	0.00	0.30	52.23	151.92
L20xT1	-1.21	-20.29	-27.48	0.35	2.36	1.94	0.26	1.22	1.50	0.14	-6.07	55.45
L20xT2	1.21	7.63	-2.08	-0.35	0.00	-0.41	-0.26	-0.95	-0.68	-0.14	-18.46	34.93
L21xT1	0.54	-8.57	-16.83	-0.48	3.75**	3.32*	-0.85	3.39*	3.67*	0.14	-35.12	7.38
L21xT2	-0.54	-21.94	-28.98	0.48	3.47**	3.04*	0.85	3.93**	4.22**	-0.14	-46.74	-11.86
L22xT1	-1.24	-15.85	-23.45	0.58	2.22	1.80	0.81	2.17	2.45	-0.7	21.82	101.61
L22xT2	1.24	12.76	2.58	-0.58	-0.69	-1.11	-0.81	-1.36	-1.09	0.7	108.07*	244.34**
L23xT1	1.18	15.61	5.18	0.13	2.92*	2.49	-0.3	2.44	2.72	-0.1	121.76*	266.99**
L23xT2	-1.18	-12.79	-20.66	-0.13	1.11	0.69	0.3	1.63	1.90	0.1	138.22**	294.23**
L24xT1	-0.02	-3.59	-12.29	-0.65	1.67	1.24	-0.58	1.49	1.77	-0.21	58.23	161.86*
L24xT2	0.02	-3.76	-12.44	0.65	1.81	1.38	0.58	1.36	1.63	0.21	86.83	209.19**
L25xT1	0.26	-11.53	-19.52	-0.42	2.08	1.66	-0.69	1.09	1.36	-0.10	56.10	158.34*
L25xT2	-0.26	-18.12	-25.51	0.42	1.67	1.24	0.69	1.22	1.50	0.10	72.17	184.93*
SE	0.97	1.45	1.45	0.8	1.06	1.06	0.93	1.22	1.22	0.57	0.81	0.81
SE (Sji-Skl)	1.37			1.13			1.28			0.81		

*=0.05 and **= 0.01 significant probability level. SCA = specific combining ability, SH = standard heterosis, SE = standard error, SE (sji-Skl) = standard error of the difference of specific combining ability effects of line by testers.

standard heterosis for grain yield, some crosses showed positive heterosis over both standard checks. Maize hybrids that perform better than the checks could be used for release as hybrid variety after re-evaluation in multi-location trials. Generally, the results obtained in this study could be helpful to design appropriate breeding strategy for developing QPM hybrids and synthetics adapted to the mid altitude sub-humid agro-ecologies of Ethiopia.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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ABBREVIATIONS

BNMRC, Bako National Maize Research Center; **CM,** conventional maize; **EIAR,** Ethiopian Institute of Agricultural Research; **GCA,** general combining ability; **LSD,** least significant difference; **masl,** meters above sea level; **QPM,** quality protein maize; **SCA,** specific combining ability.

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Upland rice response to fertilizer in three agro-ecological zones of Uganda

Kayuki C. Kaizzi[1*], Angella Nansamba[1], Ruth Kabanyoro[2], Jimmy Lammo[3] and Harrison Rware[4]

[1]National Agricultural Research Laboratories, (NARL), P. O. Box 7065, Kawanda, Kampala, Uganda.
[2]Mukono Zonal Agricultural Research and Development Institute (MuZARDI), P. O. Box 164, Mukono, Uganda.
[3]National Crops Resources Research Institute (NaCRRI), P. O. Box 7084, Kampala, Uganda.
[4]CAB International (CABI), P. O. Box 633-00621, Nairobi, Kenya.

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The yield of upland rice (*Oryza* spp.) per unit area of production in Uganda is low, partly due to low soil fertility, and use of fertilizer and manure is also low. Trials to establish the response of upland rice to applied nutrients and economically optimal nutrient rates (EOR) were determined at Tororo, Kawanda and Kadesok. The increase in paddy yield was in the range of 91 to 173% with application of 50 to 120 kg N ha^{-1}. Application of 10 kg P ha^{-1} and 20 kg K ha^{-1} resulted in a 10 to 46% additional increase, respectively. Nitrogen application was profitable for all cost of fertilizer to farm-gate price of produce (CP) ratios, with a three season mean economically optimum nutrient rate (EONR) ranging from 67 to 144 kg ha^{-1} and CP ratios varying from 2 to 12. Application of 10 kg P and 20 kg K ha^{-1} together with N reduced the mean EOR to a range of 47 to 97 kg ha^{-1} depending on the CP ratio. Application of Zn, S, B and Mg together with nitrogen, phosphorus and potassium fertilizer (NPK) increased paddy yield by 19% above the NPK yield of 3.7 t ha^{-1}, indicating that either Mg, S, Zn and B or their combination limit rice production in Uganda. Trace elements were applied as a mixture. There is a need to establish which element limits rice production in addition to the economics of their use. This information is required for fertilizer blending in the region to produce blends of the right formulation. The cost of fertilizers increases with the nutrients applied, which has an implication for the appropriate quantity of fertilizers to be used. To reduce or stop soil degradation from nutrient mining requires interventions at policy level such as fertilizer subsidies, improved market for produce and input supply efficiency, and increased access to extension, information and credit by farmers.

Key words: Economic, fertilizer use, nitrogen, phosphorus, potassium, trace elements, secondary elements.

INTRODUCTION

Upland rice production in Uganda has increased substantially due to high price offered but production equals about 75% of consumption (Government of the Republic of Uganda, 2009). The increased production has been achieved more through increased production area when compared with increased yields (WARDA, 2007). Average grain yield in Uganda is estimated to be 1.5 t ha^{-1} (FAOSTAT, 2011). Inadequate control of numerous constraints including biotic and abiotic, low use of inputs, and low inherent soil N and P contribute to the low yield (Mghase et al., 2010). Little, if any, fertilizer or manure is used for upland rice production as also for

Table 1. Selected physico-chemical characteristics of the soils at Tororo DATIC, Kawanda and Kadesok.

Site-Season[†]	**pH**	**Organic matter**	**P**	**K**	**Sand**	**Clay**	**Silt**	[‡]**Textural Class**
		g kg^{-1}	**mg kg^{-1}**		**g kg^{-1}**			
Season 2013^{b}								
Kadesok	6.0	18	2.3	182	750	179	71	SL
Kawanda	5.9	36	4.0	315	681	167	152	SL
Tororo	5.2	28	4.3	194	617	233	150	SCL
Season 2014^{a}								
Kadesok	5.9	28	2.6	201	738	202	60	SL
Kawanda	6.0	34	6.1	290	624	200	176	SCL
Tororo	5.5	30	4.0	190	626	249	125	SCL
Season 2014^{b}								
Kadesok	6.1	22	3.9	191	811	134	56	SL
Kawanda	6.0	40	6.8	241	506	298	196	SCL
Tororo	5.8	25	4.6	174	532	334	133	SCL

[†] The coordinated and elevation of the research locations, respectively were: Kawanda, 0.411567, 32·53621, 1172 m, Petric Plinthsol; Kadesok, 1·126259, 33.86942, 1079 m, Acric Ferralsols Plinthsol; and Tororo, 0.66518, 34.19825 1207 m, Petric Plinthsol. SCL, sandy clay loam; SL, sandy loam.

other crops in Uganda, as well as in other sub-Saharan African countries, due to high costs of fertilizer use relative to the price of rice (Gitau et al., 2011) plus other socioeconomic constraints. Unfortunately, soil nutrient mining is high and a major cause of land degradation in Uganda. The estimated mean depletion rates for nitrogen, phosphorus and potassium are -21, -8 and -43 kg ha^{-1} year^{-1}, respectively, in Uganda (Wortmann and Kaizzi, 1998).

Research findings indicate that upland rice respond to fertilizers with grain yield increase of more than 100% with application of N and P, to *Azolla* spp. and to a preceding *Mucuna pruriens* L. green manure crop (Kaizzi et al., 2007). Yield increases in the range of 2.1 to 5.2 t ha^{-1} in response to application of 80 to 120 kg N ha^{-1} were reported in Uganda (Onaga et al., 2012). Paddy yield could be increased by 46 kg ha^{-1} per 1 kg ha^{-1} of applied N (Miyamoto et al., 2012). In the Ivory Coast, maximum yield was obtained with 50 kg ha^{-1} of NPK fertilizer 12:24:18 or with 12 kg ha^{-1} of urea-N applied. The low response was due to soil water deficit stress during grain fill (Galabi et al., 2011). Upland rice yields were increased from 1.7 to 2.3 t ha^{-1} with a Bray-1 soil test P of 4 mg kg^{-1}, and with appropriate P application (Oikeh et al., 2010); and from 0.98 to 1.27 t ha^{-1} with Bray^{-1} of 2 to 3 mg kg^{-1} (Sahrawat, 2000) with 45 kg ha^{-1} P applied. The optimum N and P application rates for upland rice production by smallholders in Nigerian forest agro-ecosystems are 60 and 26 kg ha^{-1} of N and P, respectively (Oikeh et al., 2008).

The objectives of this research were to quantify the yield response of upland rice to N, NPK, a combination of NPK, secondary and trace elements; to determine economically optimal nutrient rates for N, and for a combination of N together with 10 kg P ha^{-1} and 20 kg K ha^{-1} at different cost of nutrient (C) to farm-gate price of produce (P) [CP] ratio.

MATERIALS AND METHODS

Site characteristics and experimental design

Response trials were conducted at three sites namely: Tororo DATIC, National Agricultural Research Laboratories (NARL) – Kawanda and at Kadesok, Pallissa District located in Lake Victoria Crescent and Southern Lake Kyoga Basin Agroecological Zones, respectively (Wortmann and Eledu, 1999). The rooting depth of the soils was over 1.0 m. Composite soil samples from 10 cores were collected from 0- to 20-cm depth, before planting and fertilizer application. They were use in determining selected physico-chemical properties. Particle size distribution was determined according to Bouyoucos (1936), soil organic matter by Walkley and Black (1934), and available P by Mehlich 3 (Mehlich, 1984).

Soil properties varied across the research sites (Table 1). The soils at Kawanda were relatively more fertile than those of Tororo and Kadesok using soil fertility criteria established by Foster (1981). The soil organic matter levels on average ranged between 23 and 37 g kg^{-1} at Kadesok and Kawanda, respectively. The Mehlich-3 P

*Corresponding author. E-mail: kckaizzi@gmail.com. Tel: +256772518875. Fax: +256414567649.

at all sites was very low and exchangeable K was adequate. The soils are mainly Acric Ferralsol and Petric Plinthsol, with N and P availability closely related to soil organic matter content and presence or absence of fallow periods (Foster, 1981; Jones, 1972).

Experimental design

The trial design was a randomized complete block design with treatments replicated three times. The N rates evaluated were 0, 30, 60, 90, 120; the P–K rates evaluated were: 10–20; and N-P-K rates were 30–10–20, 60–10–20, 90–10–20, 120-10-20 60–20–20, 60–30–20, 60–10–40 and 60–10–60 kg ha^{-1}. The N-P-K-S-Zn-Mg-B applications for diagnostic treatment were 60-10-20-33-2.5-10-0.5. The number of treatments were limited by the incomplete factorial arrangement in consideration of Liebig's law of the minimum, proposed by J. von Liebig in 1840, expecting N to be the most limiting, followed by P, and K the least limiting major nutrient deficiencies. The N_0 treatment was used as a control. However, the P and K effects were tested only with N applied. There was confounding of P and K treatments. The size of the plots was 4.2 m by 6 m.

Urea, triple super phosphate and potassium chloride were the N, P and K sources, respectively. The S, Zn, Mg and B sources were $MgSO_4$, $ZnSO_4$ and granular boron. Phosphorus fertilizer P was applied pre-plant, N and K fertilizers were applied in three splits with 25% pre-plant, 25% at tiller formation, and 50% at panicle initiation. At planting, the fertilizers were surface broadcast and incorporated. The side dress application of N and K was band-applied to the side of the row and covered immediately with soil.

Crop management and data collection

The fields were prepared using disk plow at 15 to 20 cm depth followed by secondary disk tillage at 10 cm depth. Seeds were planted at a spacing of 20 cm by 20 cm to give a final plant population of 50 plants m^{-2}. Weeds were controlled by weeding with hand hoes twice or thrice depending on weed intensity. Chloropyrifos 5% (Dursban^{TM}) was applied for control of the stem borer complex and the African rice gall midge.

At harvest, plants from the inner rows in a 1.5 x 2.0 m area were cut at ground-level and air dried for at least 3 days. The panicles were threshed and the remnants were added to the straw and weighed together to determine the straw yield. The harvested grain was weighed, and grain yield calculated. Paddy yield was adjusted to 140 g kg^{-1} water content.

Data analysis

Data satisfied the assumptions of ANOVA. The data analyses were done by site-season using Statistix 10 (Analytical Software, Tallahassee, FL) with replications as random variables and varieties and nutrient application rates as fixed variables. When significant nutrient rate effects occurred, an asymptotic yield function was determined: Yield (t ha^{-1}) = $a - bc^N$, where a is maximum yield, b is maximum gain yield due to application of the nutrient, and c^N determined the shape of the curvilinear response, where c is a curvature coefficient and N the nutrient rate. Upland rice response to applied N was determined with and without 10 and 20 kg ha^{-1} P and K, respectively, applied.

The nutrient application rates (EOR) that gave the greatest net return ha^{-1} to fertilizer application, were calculated for a range of cost of nutrient (C) to farm-gate price (P) of produce ratio, that is, CP ratio. The CP ratios used were in the range 2 to 12. A paddy price of US$ 0.53 kg^{-1} (exchange rate of Uganda Sh. 3400 per US$) was used for the economic analysis. Equations were developed using cost of nutrient to farm-gate price of produce ratio of 2:12, non-linear regression analysis was used to relate EOR to CP ratio. Differences and relationships were considered significant at P ≤ 0.05.

RESULTS AND DISCUSSION

Site characteristics

Rainfall distribution

The cumulative rainfall during the three site years is presented in Figure 1. The Analysis of variance (ANOVA) summary is presented in Table 2.

Paddy yield response to nitrogen, phosphorus and potassium

The observed response of paddy yield to N application was consistent with results from earlier research (Kaizzi et al., 2007; Onaga et al., 2012). The mean maximum paddy yield of 3.8 t ha^{-1} was obtained with application of 120 kg ha^{-1} N as compared to 1.4 t ha^{-1} with no N (N = 0 kg ha^{-1}) applied (Figure 2). Results presented in Figure 2 indicate that paddy and straw yield were affected by N rate and N x PK interaction but not by 2- or 3-way interactions of N with location or year. The N x PK interaction was due to a greater response to N with 0PK when compared with 10P20K, but with each N practice reaching a plateau at similar yields. The yield response of paddy to applied N is:

$$Y = 3.856 - 2.474(0.977^N) \text{ With no N applied} \quad (1)$$

$$Y = 3.866 - 1.808(0.964^N) \text{ With N and (10 P and 20 K ha}^{-1}\text{) applied together} \quad (2)$$

Application of nitrogen to rice was profitable for all CPs (from 2 to 12), with Economically Optimum Nitrogen Rates ranging from 60 to 149 kg ha^{-1}, and the average for the three season ranged from 67 to 144 kg ha^{-1} depending on the CP (Figure 3). The high farm gate price of rice when compared with maize and sorghum results in relatively lower CPs and higher EONRs for rice. The significantly higher paddy yield with NPK, as compared to corresponding N treatment, confirmed that P and K limit rice production too. Though, phosphorus and potassium were applied together, the response is most likely due to phosphorus, because it is one of the most limiting nutrients in Uganda. According to Liebeg's law of the minimum, the plant will first meet its N requirement, then P and thereafter K. The significant increase in paddy yield in response to P applied together with N is consistent with results reported by Oikeh et al. (2008) and Sahrawat (2000).

The mean EONR was reduced to the range of 47 to 97

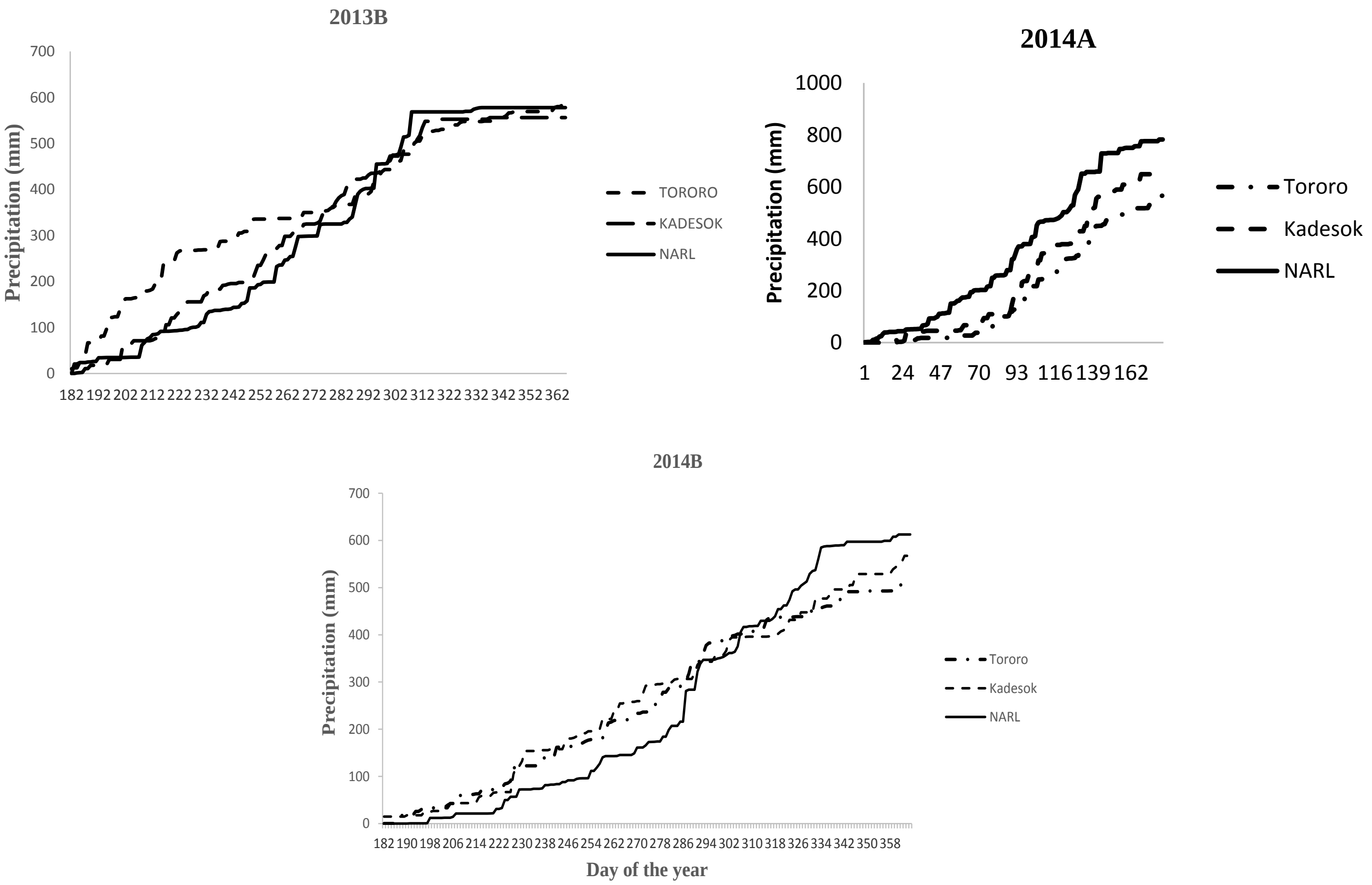

Figure 1. Cumulative rainfall at NARL, Kadesok and Tororo for the three cropping seasons.

Table 2. Analysis of variance (ANOVA) summary.

Source of variation	Degrees of freedom	Pr	
		Grain	Stover
Site year/season (S)	2	****	****
Location (L)	2	****	****
N	4	****	****
P	1	****	****
S*l	4	****	****
N*P	4	**	***
S*N	8	ns	ns
S*P	2	ns	ns
L*N	8	ns	ns
L*P	2	ns	ns
S*L*N	16	ns	ns
S*L*P	4	ns	ns
S*L*N*P	32	ns	ns
Error	180		
Total	269		
Grand mean		3.1197	8.1563
CV		12.97	15.95

ns, not significantly different at α ≤ 0.05; ** significantly different at α ≤ 0.01; *** significantly different at α ≤ 0.001; **** significantly different at α ≤ 0.0001.

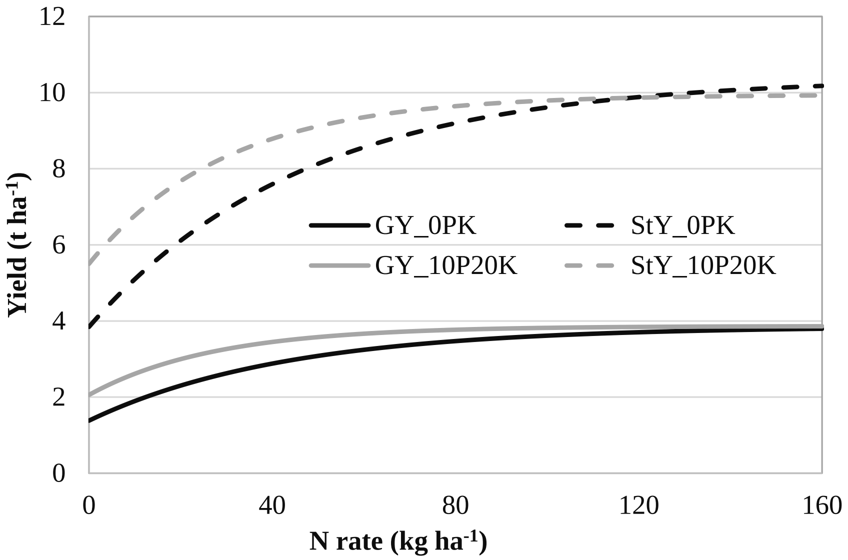

Figure 2. Upland rice response to N, with and without P and K, over nine site-seasons in Uganda.

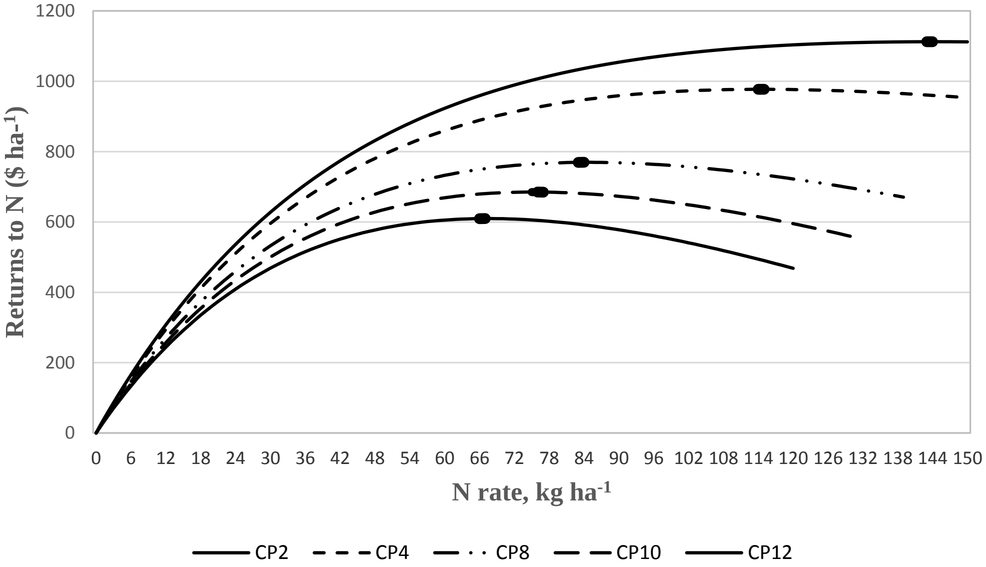

Figure 3. The average net returns for application of fertilizer N to upland rice at varying N rates and at five N cost to grain price ratios (CP), the economically optimum N rates are indicated by the symbols at the peak of the curves.

kg ha^{-1} with application of a combination of 10 kg P and 20 kg K ha^{-1} and N depending on the CP ratio which were in the range of 2 to 12. The CP of fertilizer use must include fertilizer procurement and application costs, the interest rate or opportunity cost of the money used for fertilizer purchase, and the nutrient price. These added costs are very high in Uganda and other sub-Saharan African nations (SSA) because fertilizer is not easily available (Sanchez, 2002). Yet according to CIMMYT (1988), the opportunity cost for resource-poor farmers who have little access to money or credit is often 100% of the actual value due to other high priority needs for the available funds plus other investment opportunities. A BC ≥1 is therefore required for such an investment to be attractive to the finance-constrained farmers (Wortmann and Ssali, 2001). The value of grain or paddy used in

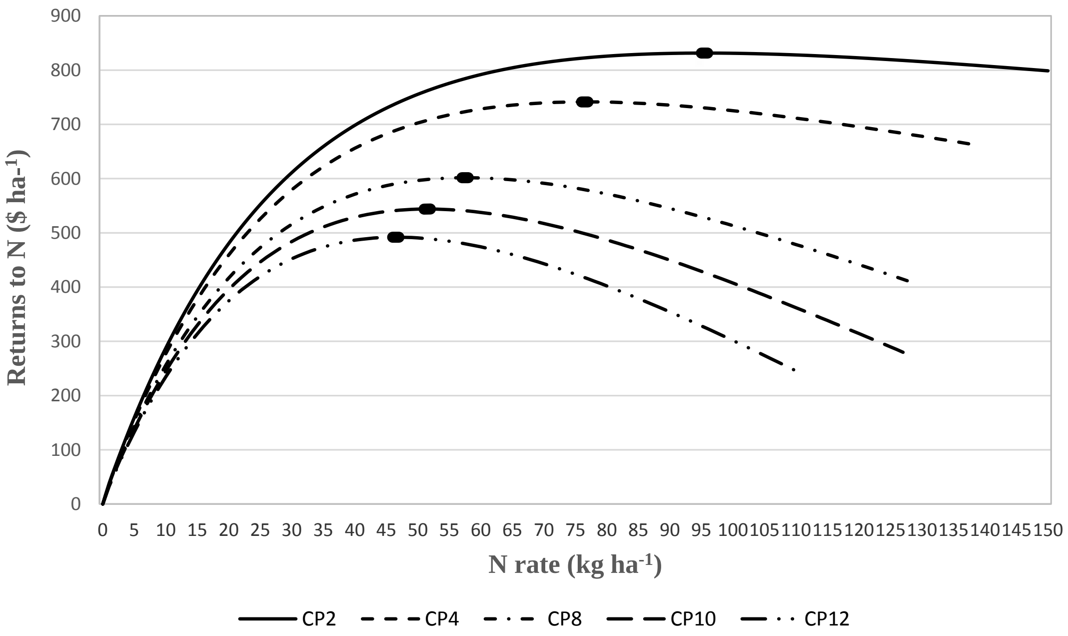

Figure 4. The average net returns of fertilizer N applied together with 10 kg P and 20 kg K per ha on upland rice, at varying N rates and at five N cost to grain price ratios (CP), the economically N optimal rates are indicated by the symbols at the peak of the curves.

determining CP, whether used for consumption by the producers or marketed, must consider the added costs of harvesting, processing, storage and marketing the increased production.

Smallholder farmers in Uganda like other SSA countries do not have enough money to purchase fertilizers for application at EOR; therefore, they maximize net returns ha^{-1}. The only option farmers have is to apply fertilizers at less than EOR over large land which gives high total production and higher net returns when compared with application at EOR to less acreage. To maximize net returns on their constrained investment, smallholder farmers have to optimize their choice of crop-nutrient-rate combinations, while considering the CPs.

The results of this study confirm that applying N to upland rice increases farm productivity with high profitability. Improved input supply and marketing efficiency, fertilizer subsidies, and improved access to credit could greatly reduce the cost of fertilizers or nutrient, hence the CP and therefore the BC of fertilizer use for upland rice production.

Net return on investment of fertilizers

The amount of money invested on nitrogen applied alone, and in combination with 10 kg P and 20 kg K ha^{-1}, are presented on the x-axis and the net returns to investment are presented on the y-axis of Figure 4. The curves show the profit potential of a nutrient applied to a crop in a given season and also the average of three seasons. The steepness of the curve indicates the level of net returns to investment, with high returns if the curve is steep. The slope decreases with increased rates of application; however, profit increases if the curve continues upward and vice versa. The peak of the curve is the point of maximum profit per unit area, referred to as the EOR.

There is seasonal variation in the profitability of fertilizer use as observed in Figure 5. This is due to fluctuations in weather and variable costs including the price of fertilizers and produce. It is important that farmers get adequate information and on a timely basis so that they can adjust based on EONR. Vlek (1990) reported that the profitability of fertilizer use in SSA is highly variable and depend on agro-climatic and economic conditions at local and regional levels. Applying fertilizers to replenish nutrients removed from the soil, to supply amendment nutrients, and also to reverse soil degradation caused by nutrient mining through soil erosion and limited/and or non-use of external inputs, require policy interventions. These interventions should include targeted fertilizer subsidies, improved market and storage facilities for produce, input supply efficiency, and/or increased access to information and credit to reduce the cost of fertilizers, hence, the cost of nutrient: farm-gate price of produce ratio (CP). The most important thing is to reduce fertilizer cost so that smallholder farmers with little money can apply fertilizers over larger acreage of their land and on several crops. Seasonal variation in the profitability of fertilizer use requires that farmers should have access to

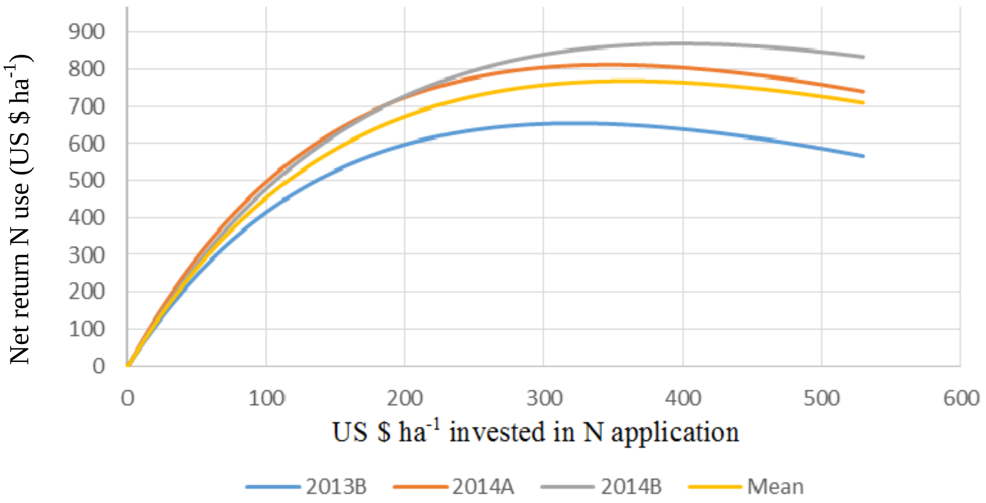

Figure 5. Net returns on investment in fertilizer N.

Table 3. Paddy yield response to N application as compared to N + (10 kg P + 20 kg K) ha^{-1} [NPK] at the different sites.

Season	N		NPK	
	Grain	Straw	Grain	Straw
	^{a}t ha^{-1}			
DATIC	2.96^{a}	7.95^{a}	3.36^{a}	8.51^{ab}
Kadesok	2.73^{b}	7.27^{b}	3.13^{b}	8.18^{b}
NARL	3.05^{a}	8.04^{a}	3.49^{a}	9.00^{a}
Pr	****	****	***	*

* Significantly different at α ≤ 0.05; *** significantly different at α ≤ 0.001; **** significantly different at α ≤ 0.0001.

more accurate and reliable weather forecasts and stable grain prices to estimate better EONR for the season.

Seasonal and site effect on response of rice nitrogen, phosphorus and potassium

There was significant mean paddy rice yield response between sites (Table 3). This is attributed to differences in selected physico-chemical characteristics of the soil presented in Table 1.

Effect of Zn, S, Mg, B and NPK as compared to NPK

Application of secondary and trace elements significantly increased paddy and straw yield by 0.71 and 2.2 t ha^{-1}, respectively over NPK only. The nutrients in the diagnostic package were a combination of N, P, K, Mg, S, Zn and B. This implies that either Mg, S, Zn and B or their combination also limit rice production in Uganda. However, trace elements were applied as a mixture, this will require conducting nutrient omission trials to establish which element limits rice production. There is need for data on crop response to secondary and trace elements, to guide in the formulations of fertilizer blends in SSA. A number of fertilizer blending fertilities have been established across the region but with limited data. Another critical issue to be addressed is the economics of fertilizer blends, because the profitability of fertilizer use decreases with application of more nutrients.

Conclusion

Application of 50 to 120 kg N ha^{-1} increased paddy rice yield by 91 to 173% and further 10 to 46% increase with application of 10 kg P ha^{-1} and 20 kg K ha^{-1} together with N, confirming that N, P and K limit rice production in Uganda. Therefore, application of N, P and K fertilizers is effective in increasing rice yield in Uganda. This has beneficial effects on food security, a primary goal of both the smallholder farmers and Government of Uganda. It increases farmer's income through sales of surplus products, thus improving farm profitability. Fertilizer use on rice is profitable as observed from economic analysis. Nitrogen application was profitable for all CPs, with three-season mean EONR ranging from 67 to 144 kg ha^{-1} with CP ratios varying from 2 to 12. The EONR were reduced to the range 47 to 97 kg ha^{-1} depending on the CP ratio when N was applied together with 10 kg P and 20 kg K ha^{-1}; this is due to the increased cost of fertilizers. The profitability of fertilizer use varies per season due to changes in variable costs and the price of produce. Therefore, farmers should adjust EONR based on current information on variable costs and anticipated produce price at the end of the season. The EOR for rice are relatively high as compared to maize and sorghum, due to the fairly high price of rice. The EOR are sufficient to reduce soil nutrient depletion but inorganic fertilizers should be used together with organic materials to derive the synergism between the two.

Application of Zn, S, B and Mg together with NPK resulted in an increase of 19% in grain yield above the NPK yield (3.67 t ha^{-1}), indicating that either Mg, S, Zn and B or their combination are limiting yield in Uganda. Since trace elements were applied as a mixture, there is a need to conduct nutrient omission trials to establish which element limits rice production. The information is required by the fertilizer blending facilities established in the country to produce the right formulation; but in the region that is in SSA due to the mushrooming blending facilities, this will prevent exploitation of the smallholder farmers by the manufacturers. The economics of fertilizer blends should be determined, since the profitability of fertilizer use decreases with application of more nutrients. To reverse soil degradation due to nutrient mining, there is need for interventions at some policy level to reduce the CP ratios. These include but not limited to targeted fertilizer subsidies, improved markets for produce and input supply efficiency, collective marketing, better and timely access to information by farmers and affordable credit. The most important issue is to reduce fertilizer cost to enable poor farmers apply fertilizer to larger acreage and on different crops.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

ACKNOWLEDGEMENTS

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Full Length Research Paper

Genetic variability, correlation and path analysis for quantitative traits of seed yield, and yield components in chickpea (*Cicer arietinum* L.) at Maichew, Northern Ethiopia

Assefa Amare Hagos[1]*, Tadesse Desalegn[2] and Tesfay Belay[3]

[1]Ethiopian Institute of Agricultural Research, Mehoni Agricultural Research Center, Maichew, Tigray, Ethiopia.
[2]Department of Plant Science, College of Agriculture and Environmental Science, Bahir Dar University, Bahir Dar, Ethiopia.
[3]Tigray Agricultural Research Institute, Mekelle Agricultural Research Center, Mekelle, Ethiopia.

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Twelve chickpea genotypes were tested to assess variability, heritability, correlations and direct and indirect effects between yield and yield components. Maximum phenotypic and genotypic coefficient of variation was recorded for number of seeds per plant (33.8, 32.4), number of secondary branches per plant (30.3, 29.6), number of pods per plant (25.6, 24.7) and 100 seed weight (23.0, 22.7) respectively . High heritability coupled with high expected genetic advance as percent of mean were estimated for number of secondary branches per plant, number of pods per plant and 100 seed. Path coefficient analysis (seed yield as a dependent variable) revealed that seeds per plant followed by biomass yield, days to maturity and 100 seed weight had exerted positive direct effect on seed yield. To conclude, number of seeds per plant, biomass yield, 100 seed weight and days to maturity are important parameters for selecting maximum yielding genotypes in chickpea.

Key words: Chickpea, genetic variability, path coefficient, heritability, correlation, genetic advance.

INTRODUCTION

Chickpea ranks third among pulses, and it accounts for 12% of the world pulses production (Khan and Khan, 2011). In Ethiopia it accounts for about 14.31% (third) of the acreage and 17.28% (second) of the total production of all grain legumes grown in the country. Area of production has been increasing greatly in recent years. In the 2011 main (MEHER) season, about 232,000 ha of cultivated land is used for the production of 400,200 tons of chickpea (CSA, 2012). Chickpea, a multi-functional crop, has an important role in the diet of the Ethiopian small scale farmers' households and also serves as protein source for the rural poor who cannot afford to buy animal products. The crop also serves as a source of cash income and plays a major role in Ethiopia's foreign

*Corresponding author. E-mail: assefaamare69@yahoo.com. Tel: +251-914-297477. Fax: +251347770021.

exchange earnings through export to Asia and Europe. Despite its nutritional values and economic importance, the average yield of chickpea is relatively low in the country. This is primarily due to poor genetic makeup of the cultivars available, excessive vegetative growth, low tolerance to diseases and non-availability of grains of improved varieties which need immediate attention of the breeders for the evolution of maximum yielding varieties which fulfill the requirements of ever increasing population.

Genetic variability is a prerequisite for any breeding program, which provides opportunity to a plant breeder for selection of high yielding genotypes. However information on the association between yield and its various components provide the basis for the selection of improved varieties (Saleem et al., 2005). Information on the relative magnitude of the different sources of variation particularity among different genotypes for several traits helps in measurement of their range of genetic diversity and may provide evidence for identification of their relationship. The variability of a biological population is an outcome of genetic constitution of the individuals and its interaction with the prevailing environment. A survey of genetic variability with the help of suitable parameters such as genetic coefficient of variation, heritability estimates and genetic advance are absolutely necessary to start an efficient breeding program. Some of the characters are highly associated among themselves and with seed yield. The analysis of the relationships among these characters and their associations with seed yield is essential to establish selection criteria (Atta et al., 2008). Progress in any breeding program depends upon the nature and magnitude of variability present in the base population. Assessment of the extent of genetic variability within chickpea is fundamental for chickpea breeding (Qureshi et al., 2004).

Chickpea breeders should consider heritability estimates along with genetic advance because heritability alone is not a good indicator of the amount of usable genetic variability (Noor et al., 2003). The concept of heritability explains whether the differences observed among individuals arose as a result of differences in genetic makeup or due to environmental forces. Genetic advance gives an idea of possible improvement of new population through selection, when compared to the original population. The genetic gain depends upon the amount of genetic variability and magnitude of the masking effect of the environment. Information of the genetic variability, heritability and association of various characters provides a basis to the plant breeders to breed the chickpea genotypes possessing higher yield potential. Selection on the basis of grain yield, a polygenic character, is usually not very efficient, but selection based on its component characters could be more efficient. The present study was initiated with the prime objective of finding the mutual relationships of different quantitative traits and the type and extent of their contribution to grain yield.

MATERIALS AND METHODS

Description of the experimental site

The field experiment was conducted in Maichew Agricultural College which is located at 39°32´E and 12°47´N in the Tigray National Regional State, Ethiopia. Maichew is found 123 km far from mekelle the capital city of Tigray region and 662 km north of Addis Ababa the capital city of Ethiopia. It is located at 2396 m above sea level and receives an average annual rain fall of 758.7 mm and annual mean temperature of 16.4°C.

Experimental materials and procedures

Twelve chickpea genotypes obtained from the High Land Pulse Research Program of Debre Zeit Agricultural Research Center (Table 1) were planted in a randomized complete block design with three replications. Each plot consisted of 4 lines of 4 m length by 1.2 m width (4.8 m^2). The plant-to-plant and row-to-row distance was maintained at 10 and 30 cm, respectively. Agronomic practices were carried out as per recommendation.

Data collection

The following data were collected from the experiment both per plot and per plant basis.

Data recorded on plot basis

a) Days to 50% flowering (DF)
b) Days to 90% maturity (DM)
c) Grain filling period (GFP)
d) Hundred Seed weight (HSW)
e) Biomass yield (Biological yield) (BY)
f) Seed yield per hectare (SY)

Data recorded on plant basis

The data for the following characters were recorded from five randomly taken plants from each plot and the average value was considered per plant basis.

a) Plant height (PH)
b) Number of Primary Branches per Plant (NPB)
c) Number of Secondary Branches per Plant (NSB)
d) Number of Pods per Plant (PPt)
e) Number of Seeds per Pod (SPo)
F) Number of Seeds per Plant (SPt)

Statistical analysis

The phenotypic, genotypic and environmental variances and coefficient of variation is defined according to the formula suggested by Singh and Chaudhary (1985) as follows:

$$Environmental\ variance\ (\sigma^2 e) = MS_e$$

$$Genotypic\ Variance(\sigma^2 g) = \left[\frac{MS_g - MS_e}{r}\right]$$

Table 1. List of genotypes considered in the study.

S/N	Variety	Year of release	Crosses/seed source	Seed color	Type
1	DZ-10-11	1974	Collection	Light Brown	Desi
2	Dubie	1978	Collection	Grey	Desi
3	Mariye	1985	K850xF378(sel fromICCx730089)	Brown	Desi
4	Wroku	1994	(Annigeri x Chaffa) x (Rabat xF378)	Golden	Desi
5	Akaki	1995	P99 xNEC 108) x Radhey	Golden	Desi
6	Mastewal	2006	NA	Golden	Desi
7	Naatolii	2007	(ICCV88102 x ICCV10) x ICC4958	Light Green	Desi
8	shasho	1999/2000	ICCC33x(L144xE100Y(M)	White	Kabuli
9	cheffe	2004	(ICCV2xsurutato 77)xICC7344	White	Kabuli
10	Habru	2004	X85TH230/ILC3395xFlip83-13C	White	Kabuli
11	Ejeri	2005	X94TH71/FLIP87-59CxUC15	White	Kabuli
12	Teji	2005	X94TH75/FLIP87-58CxUC15	White	Kabuli

$$Phenotypic\ variance (\sigma\, p^2) = \sigma g^2 + \sigma e^2$$

$$Phenotypic\ Coefficient\ of\ variation\ (PCV) = \frac{\sigma p}{\bar{x}} x100$$

$$Genotype\ coefficeint\ of\ variation\ (GCV) = \frac{\sigma g}{\bar{x}} x100$$

Where, $\bar{x}$ = grand mean of character.

Broad sense heritability (H) expressed as the percentage of the ratio of the genotypic variance to the phenotypic variance will be computed on genotype mean basis as described by Allard (1960) as:

$$H = \left[\frac{\sigma_g^2}{\sigma_p^2}\right] x100$$

Genetic advance in absolute percent of the mean (GAM), assuming selection of superior 5% of the genotypes will be estimated in accordance with the methods illustrated by Johnson et al. (1955).

GA= $K\sigma^2 P\ h^2$

GMA = (GA/$\bar{x}$) / x 100

Phenotypic and genotypic correlation coefficients were estimated using the standard procedure suggested by Miller et al. (1958) from corresponding variance and covariance:

$$phenotypic\ correlation\ coefficient\ \left(r_{P_{xy}}\right) = \frac{\sigma_{F_{xy}}}{\left(\sqrt{\sigma^2 P_x * \sigma^2 P_y}\right)}$$

$$\text{Genotypic correlation coefficient}\ \left(r_{g_{xy}}\right) = \frac{\sigma_{g_{xy}}}{\left(\sqrt{\sigma^2 g_x * \sigma^2 g_y}\right)}$$

$$t = \frac{r}{\sqrt{\frac{1 - r^2}{n - 2}}}\ where, n\ number\ of\ genotypes$$

$$t = \frac{rg_{xy}}{SE_{rg_{xy}}}\quad where, SE_{rg_{xy}} = \sqrt{\frac{1 - r^2\, g_{xy}}{2h^2{}_x * h^2{}_y}}$$

Phenotypic correlation coefficient was tested for their significance using the formula suggested by Sharma (1998).

$$t = \frac{r}{\sqrt{\frac{1 - r^2}{n - 2}}}\ where, n\ number\ of\ genotypes$$

Genotypic correlation coefficient was tested with the following formula suggested by Robertson (1959):

$$t = \frac{rg_{xy}}{SE_{rg_{xy}}}\quad where, SE_{rg_{xy}} = \sqrt{\frac{1 - r^2\, g_{xy}}{2h^2{}_x * h^2{}_y}}$$

$SErg_{xy}$ = Standard error of genotypic correlation coefficient between character X and Y

h^2x = heritability for character x and h^2y = heritability for character y. The calculated absolute t value was tested against the tabulated t-value at g-2 degree of freedom for both phenotypic and genotypic correlations.

Path coefficient analysis was estimated as suggested by Dewey and Lu (1959) using the phenotypic as well as genotypic correlation coefficients to determine the direct and indirect effects of yield components on seed yield based on the following relationship:

$$\mathbf{r_{ij} = P_{ij} + \Sigma\, r_{ik}\, P_{kj}}$$

Where, **rij** = Mutual association between the independent character (i) and dependent character, grain yield (j) as measured by the correlation coefficients. **Pij** = Components of direct effects of the independent character (i) as measured by the path coefficients and $\mathbf{\Sigma\ r_{ik}\ p_{kj}}$ = summation of components of indirect of a given independent character (i) on a given dependent character (j) via all other independent characters (k). The contribution of the remaining unknown factor was measured as the residual factor (PR), which is calculated as:

Table 2. Genetic parameters of yield and yield components in chickpea.

Trait	GV	PV	EV	PCV (%)	GCV (%)	H (%)	GA	GA (%)
DF	47.78	47.30	0.47	14.64	14.56	99	14.10	29.85
DM	11.74	10.20	1.53	2.98	2.78	87	6.13	5.34
GFP	46.18	45.28	0.90	10.06	9.96	98	13.74	20.32
PH	17.49	15.63	1.86	13.84	13.08	89	7.69	25.47
NPB	0.05	0.01	0.04	8.62	2.84	11	0.10	1.93
NSB	5.30	5.05	0.25	30.28	29.56	95	4.52	59.43
PPt	97.39	90.12	7.28	25.64	24.67	92	18.62	48.88
SPo	0.02	0.01	0.01	12.00	10.11	71	0.21	17.55
SPt	235.09	216.59	18.50	33.80	32.44	92	29.09	64.15
BY	230991.52	205172.36	25819.15	14.15	13.34	87	879.31	25.90
SW	42.29	41.20	1.20	23.03	22.70	97	13.02	46.10
SY	115361.95	104073.23	8.73	16.0	16.00	90	631.01	31.30

DF = Days to 50% flowering, DM = Days to maturity, GFP = Grain filling period, PH = Plant height (cm), number of primary branches per plant, NSB = Number of secondary branches per plant, PPt = Number of pods per plant, SPo= Number of seeds per pod, SPt = Number of seeds per plant, BY = Biomass yield (kg/ha), SW = 100 seed weight (g), SY= Seed yield (kg/ha) , SE=Standard error, GV= Genotypic Variance, PV=phenotypic Variances, EV=Environmental Variance, PCV= phenotypic of variability, GCV= genotypic coefficient of variability, H=broad sense heritability, GA= expected genetic advance GA% =genetic advance as percent of the mean GA%.

$$p_r = \sqrt{\left(1 - \sum r_{ij} P_{ij}\right)}$$

The magnitude of PR indicates how best the causal factors account for the variability of the dependent factor (Singh and Chaudhary, 1999).

RESULTS AND DISCUSSION

It is clear from the Table 2 that The highest estimates for phenotypic coefficients of variation were recorded for number of seeds per plant (33.80), number of secondary branches per plant (30.28), number of pods per plant (25.64) and 100 seed weight (23.03). The higher phenotypic coefficients of variation values for number of pods per plant and 100 seed weight were in agreement with previous reports (Sharma and Saini, 2010). The highest genetic coefficients of variation were observed for number of seeds per plant (32.44), number of secondary branches per plant (29.56), number of pods per plant (24.67) and 100 seed weight (22.70). Similar results were reported (Sharma and Saini, 2010) who found high GCV values for secondary branches per plant, pods per plant and seeds per plant in chickpea genotypes. heritability estimate was high (>80%) for days to 50% flowering, grain filling period, 100 seed weight, number of secondary branches per plant, number of seeds per plant, number of pods per plant, seed yield, plant height, days to maturity and biomass yield. High heritability values for 100-seed weight, number of pods per plant, seed yield per plant, number of branches per plant and plant height were in accordance with previous reports by Sharma and Saini (2010).

Genetic advance as percent of mean at 5% selection intensity was high for number of seeds per pod (64.2%) followed by number of secondary branches per plant (59.4%), number of pods per plant (48.9%) and 100 seed weight (46.1%). Ali et al. (2011) found higher values of genetic advance for number of pods per plant, plant height and grain filling period. The present study revealed that high heritability coupled with high expected genetic advance as percent of mean for number of secondary branches per plant, number of pods per plant and 100 seed weight. Therefore, these characters could be improved more easily than other characters measured in this study. Genotypic and phenotypic correlations among the characters are shown in Table 3. Seed yield showed positive and significant phenotypic association with biomass yield (0.75) and plant height (0.59) Therefore, any improvement of these characters would result a substantial increment in seed yield. Similar reports were observed by Vaghela et al. (2009), Malik et al. (2010) and Kobraee et al. (2010). The correlation coefficients of seed yield with hundred seed weight were positive at genotypic level and negative at phenotypic level. Biomass yield had significant positive genotypic and phenotypic correlation with seed yield. Similar results have been reported by Ali et al. (2011). Positive genotypic correlations of biomass yield with plant height (0.53), 100 seed weight (0.44) and number of primary branches per plant (0.32) have also been observed. Hundred seed weight had positive genotypic and phenotypic correlation with plant height. It had negative and significant genotypic and phenotypic correlation with number of secondary branches per plant ($r_{g=}$0.83, $r_{ph=}$ 0.81), seeds per pod ($r_{g=}$0.87, r_{ph}= 0.73), pods per plant ($r_{g=}$0.73, $r_{ph=}$ 0.73) and seeds per plant ($r_{g=}$0.84, $r_{ph=}$ 0.79).

Table 3. Genotypic (above diagonal) and phenotypic (below diagonal) correlation coefficients among 12 characters.

Variables	DF	DM	GFP	PH	NPB	NSB	PPt	SPo	SPt	BY	SW	SY
DF		0.295	-0.894**	-0.286	-0.454	0.190	0.223	0.137	0.195	0.159	-0.013	0.443
DM	0.299		0.163	-0.362	-0.710**	0.212	0.381	0.610*	0.569*	0.215	-0.497	0.217
GFP	-0.881**	0.186		0.132	0.104	-0.098	0.047	0.150	0.071	-0.073	-0.224	-0.358
PH	-0.263	-0.267	0.146		0.444	-0.727**	0.253	-0.322	-0.285	0.523	0.607*	0.600*
NPB	-0.169	-0.326	0.003	0.104		0.208	0.115	0.283	-0.074	0.316	0.111	0.237
NSB	0.180	0.212	-0.081	-0.641*	0.049		0.704*	0.594	0.667*	-0.538	-0.833**	-0.370
PPt	0.213	0.344	0.038	0.177	0.002	0.675**		0.618*	0.933**	-0.372	-0.727**	0.137
SPo	0.115	0.488	0.125	-0.251	0.020	0.484	0.494*		0.848*	-0.165	-0.868**	-0.032
SPt	0.184	0.523*	0.079	-0.209	-0.040	0.640*	0.911**	0.786**		-0.323	-0.836*	0.090
BY	0.148	0.214	-0.048	0.550*	0.045	-0.471	-0.275	-0.110	-0.231		0.465	0.766**
SW	0.408	-0.478	-0.226	0.582*	0.090	-0.805**	0.676**	-0.734**	-0.790**	0.444		0.335
SY	-0.014	0.156	-0.341	0.587*	0.099	-0.316	0.180	0.034	0.143	0.748**	-0.338	

*, ** Indicate significance at the 0.05 and 0.01 probability levels, respectively. DF = Days to 50% flowering, DM = Days to maturity, GFP = Grain filling period, PH = Plant height (cm), number of primary branches per plant, NSB = Number of secondary branches per plant, PPt = Number of pods per plant, SPo= Number of seeds per pod, SPt = Number of seeds per plant, BY = Biomass yield (kg/ha), SW = 100 seed weight (g), SY= Seed yield (kg/ha).

Negative association between 100 seed weight indicates a compensatory relationship between them. Pods per plant had positive and significant genotypic and phenotypic correlation with number of secondary branches per plant ($r_{g=}$0.70, $r_{ph=}$ 0.68) and seeds per plant ($r_{g=}$0.93, $r_{ph=}$ 0.91). A positive and significant genotypic and phenotypic correlation of number of pods per plant with number of secondary branches per plant agrees with the findings of Ali et al. (2011). Positive and significant genotypic and phenotypic correlation of seeds per plant with number of secondary branches per plant ($r_{g=}$0.67, $r_{ph=}$ 0.64), number of pods per plant ($r_{g=}$0.93, $r_{ph=}$ 0.91) and seeds per pod ($r_{g=}$0.82, $r_{ph=}$ 0.78) has been observed. Seeds per pod had significant positive genotypic and phenotypic correlation with seeds per plant ($r_{g=}$0.85, $r_{ph=}$ 0.79). Positive and significant correlation of number of secondary branches per plant with number of pods per plant ($r_{g=}$0.70, $r_{ph=}$ 0.68) and number of seeds per plant was observed at genotypic and phenotypic level. The positive and significant correlation of number of secondary branches per plant with number of pods per plant agrees with the findings of Malik et al. (2010). Plant height had positive and significant genotypic and phenotypic correlation with 100 seed weight ($r_{g=}$0.61, $r_{ph=}$ 0.58) and seed yield ($r_{g=}$0.60, $r_{ph=}$ 0.59), Plant height had positive genotypic correlation with biological yield and number of primary branches per plant. This is in line with the study by Ali et al. (2011) who found positive and non-significant genotypic correlation of plant height with number of primary branches per plant. Generally, positive and significant association of pairs of characters at phenotypic level and positive and high correlation at genotypic level justified the possibility of correlated response to select. The negative correlations prohibit the simultaneous improvement of those traits. Thus, correlation analysis indicated that biomass yield and plant height were found to be important yield components and these traits can be used for yield improvement in chickpea (Table 3).

Seeds per plant followed by biomass yield, days to maturity and 100 seed weight had exerted positive direct effect on seed yield. Deb and Khaleque (2005), Yucel et al. (2006) and Zali et al. (2011) reported similar results for seeds per plant. However, days to 50% flowering, grain filling period, number of secondary branches per plant, number of pods per plant, seeds per pod, plant height and number of primary branches per plant showed negative direct effect on seed yield. The high positive direct effect of 100 seed weight on seed yield was counterbalanced by its indirect effect via seeds per plant which finally resulted in positive and low genotypic correlation with seed yield. The residual (0.0315) indicates that characters which are included in the genotypic path analysis explained 96.85% of the total variation in seed yields (Table 4).

Table 4. Estimates of direct (bold diagonal) and indirect effect (off diagonal) for 12 characters.

Variables	DF	DM	GFP	PH	NPB	NSB	PPt	SPo	SPt	BY	SW	r_g
DF	**-1.56613**	0.12957	1.58404	0.01542	0.01231	-0.07272	0.0494	-0.02597	0.18214	0.13742	-0.0029	0.44
DM	-0.46273	-**0.43854**	0.28917	0.01948	0.01925	-0.08130	0.0845	-0.11574	0.53241	0.18610	-0.1145	0.22
GFP	1.40073	0.07160	**-1.77109**	-0.00711	-0.00282	0.03761	0.0105	-0.02838	0.06652	-0.06351	-0.0515	-0.36
PH	0.44859	-0.15865	-0.23398	**-0.05384**	-0.01204	0.27841	0.0561	0.06119	-0.26688	0.45355	0.1397	0.60*
NPB	0.71160	-0.31147	-0.18415	0.02391	**-0.02710**	-0.07948	0.0254	-0.05371	-0.06917	0.27385	0.0255	0.24
NSB	0.29758	0.09316	0.17405	0.03917	-0.00563	**-0.38271**	0.1562	-0.11274	0.62416	-0.46645	-0.1918	-0.37
PPt	-0.34889	0.16724	0.08403	0.01363	0.00311	-0.26960	**0.2217**	-0.11722	0.87247	-0.32212	-0.1674	0.14
SPo	-0.21437	0.26748	-0.26484	0.01736	-0.00767	-0.22738	0.1369	**-0.18976**	0.79298	-0.14330	-0.1998	-0.03
SPt	0.30499	0.24964	-0.12596	0.01536	0.00200	-0.25540	0.2068	-0.16089	**0.93528**	-0.27975	-0.1925	0.09
BY	-0.24830	0.09416	0.12978	-0.02817	-0.00856	0.20596	0.0824	0.03137	-0.30186	**0.86676**	0.1070	0.77**
SW	0.01962	-0.21814	0.39627	0.03268	0.00300	0.31884	0.1612	0.16474	-0.78225	0.40297	**0.2301**	0.33

Residual Effect =0.032.*, ** Indicate significance at the 0.05 and 0.01probability levels, respectively . DF = Days to 50% flowering, DM = Days to maturity, GFP = Grain filling period, PH = Plant height (cm), number of primary branches per plant, NSB = Number of secondary branches per plant, PPt = Number of pods per plant, SPo= Number of seeds per pod, SPt = Number of seeds per plant, BY = Biomass yield (kg/ha), SW = 100 seed weight (g), SY= Seed yield (kg/ha).

Conclusion

On the basis of these results it was suggested that pods per plant, primary branched per plant, secondary branches per plant and 100 seed weight may be given more importance while making selection for higher yield potential in chickpea.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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