Advanced Journal of Seed Science and Technology ISSN: 2812-3425 Vol. 6 (1), pp. 168-175, May, 2020.
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Full length Research paper

The Effect of Plant Population and Nitrogen Rates on Yield and Seed Quality of CML-395 x CML-202 Female Basic Seed for Production of BH-661 and BH-546 Hybrid Maize Seed at Bako, Western Ethiopia

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Accepted 19 May, 2020.

There is no recent agronomic recommendation of optimum N rates and plant populations for inbred lines were available to produce maize female basic seed. The study was conducted with objective of determining the effect of N rates and plant population on yield of CML-395 x CML-202 female basic seed. The experiment was laid in 3x6 factorial combinations of three levels (88,111 and 137 kgha^{-1}) and six levels of plant populations (61,538, 71,429, 76,923, 90,909, 83,333 and 66667 plants ha^{-1}) and replicated three times. 88 kg Nha^{-1} and 66,666 plants ha^{-1} were used as the standard checks. The highest and significant yield increase was recorded through practice of 83,333 plants ha^{-1} with 60cm x 20cm spacing followed by 90,909 plants ha^{-1}.The highest significant absolute net benefit (ANB) was also obtained under the practice of 83,333 plants ha^{-1}. Beyond this, the highest ANB was recorded under application of 137 kg Nha^{-1}. Use of 83,333 plants ha^{-1} by practicing 60 cm inter and 20 cm intra row spacing at the seed rate of 20.7 kg ha^{-1}(16 kg ha^{-1} of CML-395 and 4.7 kg ha^{-1} of CML-202) characterized by at least 90% germination capacity is the best recommended cultural practice to enhance productivity and profitability for seed producers. In addition, intensive fertilization (137 kg ha^{-1}) for commercial investors is preferable to enhance the profitability per unit area whereas use of 111 kg ha^{-1} is very much better than the previous recommendation and it is advisable for small scale seed business.

Keywords: Maize, Basic seed, Nitrogen, plant population.

INTRODUCTION

The production of high-quality hybrid maize (*Zea maize* L.) seeds is one of the foundations of successful agriculture and can comprehensively enhance the development level of modern agriculture(Li et al., 2019).This can be achieved through providing hybrid maize quality seeds for the farmers offers them with varieties containing improved genetic traits, such as high yield potential and unique trait combinations to counter diseases and adverse growing conditions (John *et al.,* 2014).This could be achieved through enhancing the seed replacement rate (SRR), which refers to the percentage of area sown out of the total area of a crop planted in the season by using certified/quality seeds other than the farm-saved seeds, has a direct correlation with the productivity of crops (Bhusal, 2017).

The desirable SRR, without which it is not possible to achieve higher productivity, for hybrid maize is 100%.

Low SRR indicates that the majority of smallholder farmers use farm-saved seeds through informal ways, which could have a significant yield penalty.

However, availability of certified seeds for hybrid maize depends greatly on quantity and quality production of a female basic seeds that should fulfill the national quality standards and implementation of appropriate Agronomic management. The field management of male and female lines to produce female basic seeds for

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hybrid production is very important and requires attention to timing of planting, appropriate plant population and fertilizer application rates, elimination of off-types, removal of tassels from the females before pollen shedding and other practices to maintain seed quality (John *et al.,* 2014).

Therefore, good seed quality of maize entirely depends on the quality and the quantity of maize basic lines. Currently, the demand of basic seed (female basic seed for production of BH661 and BH546 hybrid certified seed) is increasing from time to time since the demands of these hybrids varieties are very high. However, most of the investors are not willing to produce such kind of female basic seeds since its productivity is relatively very low while its cost of production is very high as compared to hybrid seed production.

In other words, cost of production of basic seed from inbred lines is directly related to the yield and quality of seed obtained per hectare of female parent (Castañeda, 2010). Similarly, the productivity of CML-395 XCML-202 crosses of female basic seed for BH-546 and BH661 is relatively very low (John *et al*., 2014), which may discourage many investors to produce. In addition, germination capacity is one of the main seed quality parameters and is determinat factors for production of quality seed.

Producitvity and quality of maize basic seeds may governed by biotic and abiotic factors. Some of the main constraints are poor genetic potential and management practices. Inappropriate plant population and rate of nitrogen fertilizer applications are among the major factors that may reduce its productivity and quality, particularly germination and vigor sty capacity. Usually, the plant population practiced by the producers is about 66,666 plants ha^{-1} (75cm between rows and 20cm between plants).

This practice can reduce the plant population per unit area and even lead to poor pollination and hence low yield.

However, optimum plant population has a direct correlation with yield of the crop (Zeleke *et al.,* 2018).In appropriate use of N rate can also significantly affect yield of the cross of parental lines and at the same time reduce its quality (Sabagh *et al.,* 2017).

Nitrogen plays significant role in various physiological operations of maize and hence it is critical for boosting the productivity of the crop (Sabagh *et al.,* 2017). However, there was no scientific research about differential responses of maize male and female inbred lines to different plant densities in combination with nitrogen rates which could be invaluable aid for producers in hybrid maize production.

Therefore, the objectives of this study was to determine the yield and seed quality responses of female (CML-395) and male (CML- 202) to varying plant population and nitrogen rates for production of CML-395xCML-202 cross of BH661 and BH-546 female basic

395xCML-202 cross of BH661 and BH-546 female basic seed.

MATERIAL AND METHODS

The experiment was conducted at Bako Agricultural Research Center (BARC) on station for two consecutive years (2017-2018). The study area is located in a sub-humid area of Western Ethiopia which lies at a latitude of $9^0$6'N and longitude of $37^0$9'E and at an altitude of 1650m above sea level. It has also an annual mean minimum and maximum air temperatures of 13.5 and 29.7°C, respectively. The area received an annual rainfall of 1598mm (2017) and 1162mm (2018) with maximum precipitation being received in the months of May to August (Figure 1). The soil of the area is reddish-brown, nitosol. It is an acidic soil with a pH range of4.5–5.6. The surrounding area is a mixed farming zone and is one of the most important maize (*Zea mays* L.) growing belts in Ethiopia and is predominantly continuous monocropping with low soil fertility problem that directly influences production and productivity of the cultivated crops.

Treatments

There were 18 treatment combinations of six levels of plant populations and three levels of nitrogen rates. The population treatments were indicated in the table below (Table1). Previous recommended plant populations of 66,666 plants ha^{-1} with 75 cm inter and 20 cm intra row spacing was used as standard check.

The amount of seed for both male and female inbred lines were estimated based on the plant population per ha and thousand seed of weight of the inbred lines. The average thousand seed weight of female (CLM-395) and male (CML-202) lines were 0.2567 kg and 0.2233 kg, respectively. Three levels of nitrogen rates (88, 111 and 137 kg Nha^{-1}) were another treatment factor that were arranged in factorial combination with six levels of plant population treatment. Therefore, a3x6 treatment combinations were arranged in factorial combinations with three replications.

Experimental procedures:

The experimental plots were cultivated two to three times and leveled very well before planting. The gross size of each experimental plot was 4m x 6 m = $24m^2$ and the distance between plots and blocks were 50 cm and 1.5m, respectively. Seeding rows were prepared using local material and the number of rows per plot was varied from 8 to 11as per the treatment arrangements.

The amount of seed required per plot was also calculated based on number of seeds to be sown per plot and thousand seed weight. The seed was planted in mid-June as per the treatment arrangements of the spacing

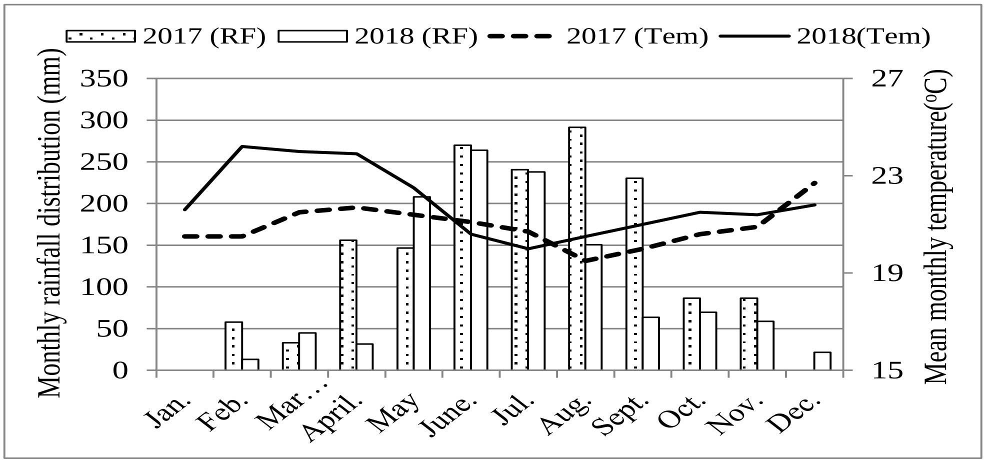

Figure1: Mean monthly temperature and monthly rainfall distribution at Bako in 2017-2018 years

Table 1: Six level of population treatments for production of CML-395xCML-202 female basic seed at Bako, 2017-2018

Spacing	Populationha^{-1}	Seed rate of inbred line (kg ha^{-1})		Total seed rate (kg ha^{-1})
		CML-395 (female)	CML-202 (male)	
65cmx25cm	61,538	11.85	3.44	15.3
70cmx20cm	71,429	13.75	3.99	17.7
65cmx20cm	76,923	14.81	4.29	19.1
55cmx20cm	90,909	17.50	5.08	22.6
60cmx20cm	83,333	16.04	4.65	20.7
75cmx20cm	66,666	12.84	3.72	16.6

between the plants. The two parental lines were planted on the same day, but the plant populations for both lines were varied as per treatment arrangements. The population ratio of female (CML-595) to male (CML-202) inbred lines for planting was 3:1, i.e 75% of the total area was covered by female inbred line to harvest CML-395 x CML-202 female basic seed for production of BH661 and BH546 certified seeds.

Phosphorus sources of 100 kg NPS (19N:38P:7S) ha^{-1} fertilizer was equally and uniformly applied to all experimental plots at the time of planting. However, nitrogen rates were applied in split application as per treatment arrangements. Half of the nitrogen (N) treatment rates were applied in the form of urea fertilizer after two weeks of seed planting whereas half of the N rates were applied in the second at the time of knee growth stage. All other agronomic managements were uniformly applied.

Data collection and analysis

Yield and yield component data were collected from the time of planting until harvesting. Stand count at emergence and harvesting time, plant height, biomass yield, grain yield, and 1000 seed weight were collected. In addition, germination percentage was measured after seed harvesting and processing.

Absolute yield response due to different plant population and nitrogen rates, which is calculated as yield obtained under various plant population treatments minus yield obtained under standard check (66,666 plants ha^{-1}) and yield obtained under various N rates minus yield obtained at recommended N rate (88 kg N ha^{-1}). Finally, the absolute yield response was adjusted downward by 10% to get adjusted absolute yield response in order to reflect the difference between the experimental yield and the yield farmers that could expected from the same treatment (CIMMYT,1988). Relative yield response was also calculated as the percentage of additional yield obtained under different plant population and nitrogen rates over the standard checks divided by yield obtained under standard checks. Finally, absolute economic benefit was calculated as adjusted absolute yield response of female basic seed multiplied by its 50 ETBkg^{-1}price (price of CML-395xCML-202 female basic seed) minus relevant absolute input cost (cost of additional inbred line seeds due to population variation and additional N rates against the standard checks as well as other additional labor cost for fertilization, shelling and threshing) following the procedure indicated in research finding (Zerihun *et.al.*,2019). The absolute variable cost was calculated based on the cost of 52.80 ETB kg^{-1} for both CML-395 and CML-202 lines seeds and cost of N rate in the form of urea is 13.54 ETB kg^{-1} and daily labor wage is 75 ETB per person for eight hours working day.

Finally, the effect of six levels of plant population/density and three levels of nitrogen rates and their interaction were analyzed by following maximum likelihood linear mixed model (REML).*Res. var= Constant + Spc + Urea + Spc.Urea + rep*; whereRes.var= responsevariate, Spc= plant spacing/plant population, Urea= sources of nitrogen rates in the form of urea, Spc.Urea= interaction effects of plant population and nitrogen rates, rep= replications. Cropping season was considered as random effect whereas plant population and nitrogen rates were considered as fixed effects. Mean separation were done using LSD value (P=0.05).Sigma plot software was used for graphing.

RESULT AND DISCUSSION

Stand count

The number of stand counts at the time of emergence and harvesting increased with an increased plant population (Figure 2). Indeed, the lowest stand count at emergence and at harvesting time was recorded with the highest plant population implying that over populated plant could significantly affected the germination capacity as the vigouristy capacity of inbred lines seeds is relatively low. A research result also shows that lower plant population of low vigor capacity of inbred lines ensures good seed set and development as it may utilize more resources than densely populated ones(Association, 2014). On the other hand, the stand count per plot was significantly lower at time of harvesting as compared to the time of emergence across various plant populations (Figure 2). Similar result also reported that plant population had significantly affected the stand counts at harvest (Imran *et al.,* 2015). This may relate to seed vigourisity capacity of the inbred line seeds which considerable reduced the potential population density.

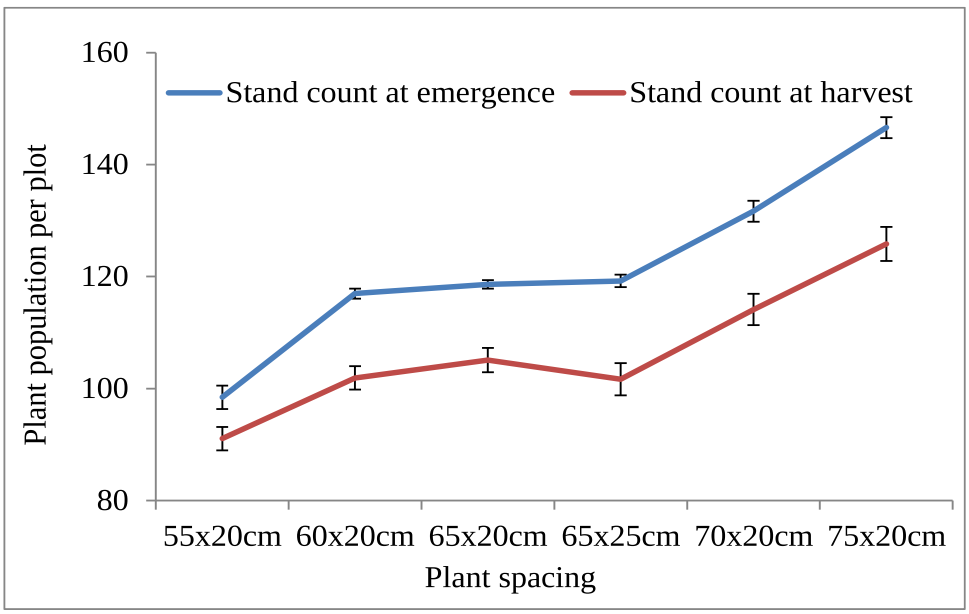

Figure 2: The effect of plant population/plant density on stand count per plot of inbred line seeds at Bako, 2017-2018

Yield and yield component

The result of analysis revealed that the main effects of plant spacing and nitrogen rates significantly affected on biomass and grain yield of female line maize seeds while plant height was significantly different by application of various nitrogen rates. On the other hand, the interaction effects of the two treatments did not show significant differences on yield and yield component of the crop (Table 2). On the other hand, thousand seed weight was significantly affected by nitrogen rates while plant population and their interaction did not show significant difference on yield and thousand seed weight.

The effect of N rate on plant height and seed yield

Plant height significantly increased by an increased N rate and the highest plant height was recorded when 111 kg N ha^{-1} was applied while the lowest plant height was

Table 2: The analysis of variance of grain yield and thousand seed weight as affected by nitrogen rate and plant spacing

		F probability			
Fixed term	Df	Plant height (cm)	Biomass (tha^{-1})	Yield (tha^{-1})	1000 seed weight(gr)
Spc	5	0.889	<0.001	<0.001	0.0117
Urea	2	0.003	0.004	<0.001	0.379
Spc.Urea	10	0.735	0.851	0.221	0.248
Rep	2	0.003	0.031	0.280	0.005

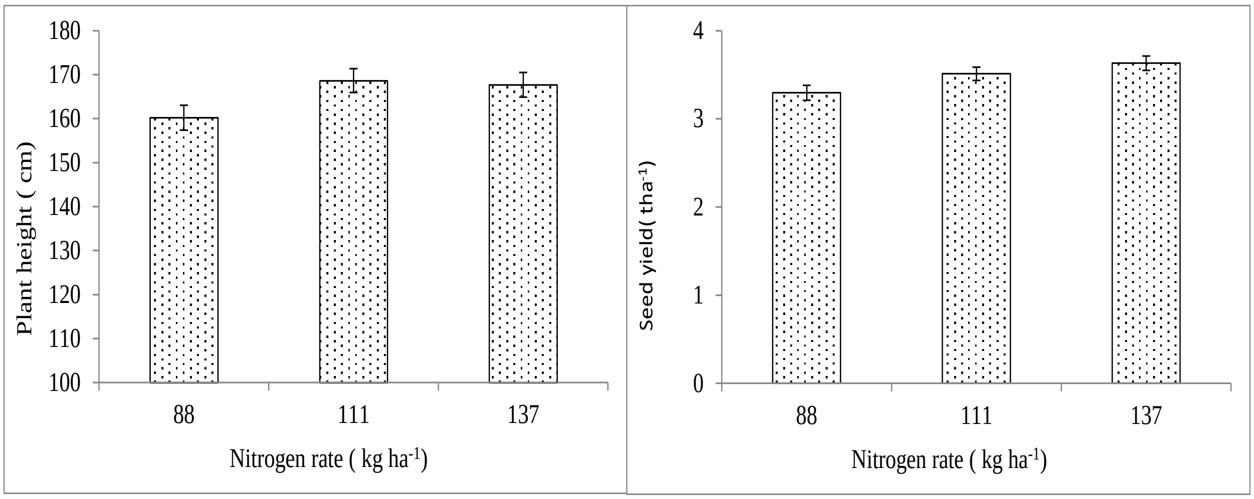

Figure 3: The effect of nitrogen rates on yield of female inbred line of maize seed at Bako, 2017-2018

recorded under minimum N application rate (Figure 3). However, the response of the crop to the highest N could lead to lodging and may also cause susceptible to pests and in turn resulted lower seed yield

The yield of female basic seed was significantly increased by 7% and 10% when 111 $kgha^{-1}$ and 137 kg ha^{-1} of nitrogen rate was applied as compared to the standard check (88 kg Nha^{-1}). In other words, more than 330 kg $ha^{-1}$of female basic seed could be harvested when the maximum N rate was applied to the crop compared to the existing N rate practice (Figure 3). Similarly, the highest biomass yield with 8% increases was obtained at the highest N application compared to the lowest N rate (data not presented). Similar result also reported in Brazil that maximum N rate (180 kg ha^{-1}) significantly increased by 3-7% of yield compared to the lowest one (60 kg Nha^{-1}) (De Faria *et al.,* 2019). This indicates that appropriate rate and time of N application to maize seed production could boost its productivity and increase marginal profit for the producers (Khodabandeh, 2010;Imran *et al.,* 2015).

The effect of plant population on biomass and seed yield

Significant differences on biomass and seed yields were observed as the function of variable plant population/plant spacing of both male and female inbred lines. The lowest biomass and seed yield were recorded at the lowest plant population (61,535 plants ha^{-1}) with 65 cm inter and 25cm intra row spacing (Figure 4).

On the other hand, the highest seed yield was obtained at 83,333plants $ha^{-1}$with 60cminter and 20 cm intra row spacing but statistically at par with yield obtained at 90,909 plants ha^{-1} (55cm x 20cm). The yield increases or relative absolute yield response (Table 3) due to use of 83, 3333 plants ha^{-1} and 90,909 plants ha^{-1}over the standard check (66,666 plants ha^{-1}) were 16% and 10%, respectively (Figure 4).

This result in line with other finding implies that intra row spacing play a significant role in determining the productivity of the crop (Abd-Alkream Hussain *et al.*, 2018; Abuzar *et.al.,* 2011;).The seeds of inbred lines need less inter and intra row spacing compared to certified seeds of hybrid maize as it is naturally less vigor and low stand development and the canopy of the leaf is also low. The yield was significantly reduced by wider intra row spacing. For instance, significantly 8% yield reduction was occurred when 25 intra raw spacing was used compared to 20 cm keeping the inter row spacing is constant (65cm). Similar result on hybrid maize yield indicates that narrow intra row spacing significantly affected on yield and yield components and the highest yield was recorded under narrow spacing (Kandil *et al.*

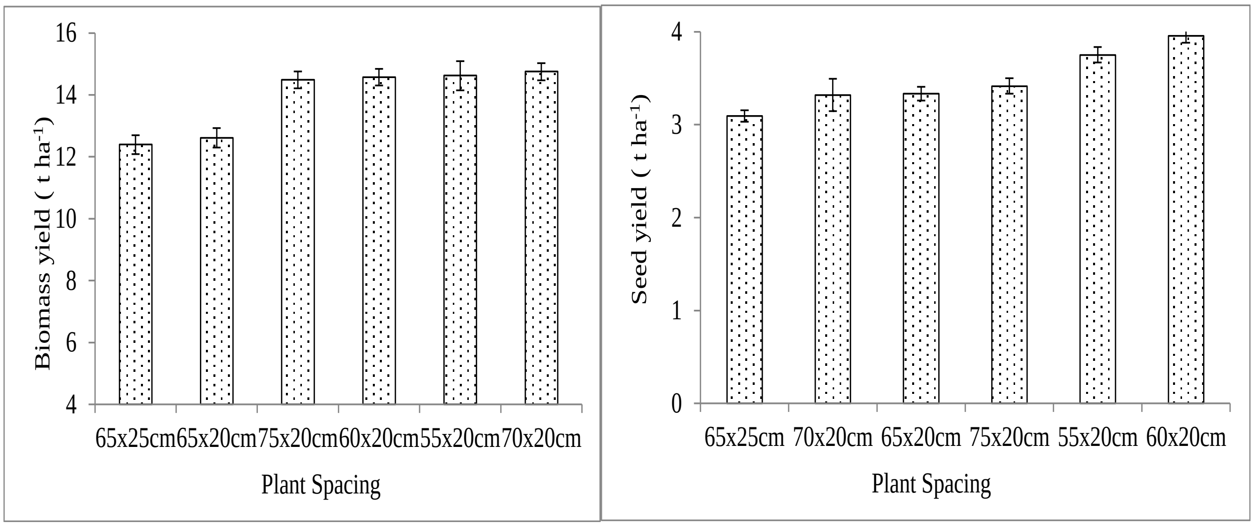

Figure 4: The effect of plant population on bimass and seed yield of basic seed of female Maize at Bako, 2017-2018

2017). On the other hand, more than 5% of yield increase was recorded under 60 cm inter raw spacing when compared to 55 cm with the same (20 cm) intra row spacing.

This implies that appropriate inter and intra row spacing for the production of basic seed of female maize seed is very important to separately use appropriate spacing to enhance the productivity of seed yield.

The productivity of female basic seed produced from cross of male and female inbred lines is relatively sensitive to intra row spacing than inter row spacing.

For instance, harvested seed yield of the crop was numerically higher (3.416 tha^{-1}) under wider inter and narrower intra row spacing (75cm x20cm) compared to narrower inter and wider intra row spacing (65 cm x 25 cm), which recorded 3.094 kg/ha seed yield.

This may be related with the leaf arrangement and canopy of the inbred lines.

Similar result also reported that plant geometry and different row arrangement can considerably influence on resource utilization to finally yield productivity (Tsuba et.al., 2001).

However, a wider inter spacing is required to produce maximum biomass yield as the crop need more spacing for higher plant canopy development.

In this case, the highest biomass yield (14.75 t ha^{-1}) was recorded at 70 cm inter row and 20 cm intra row spacing, but the yield was statistically similar at maximum and minimum plant population.

This implies that maximum plant population cause high competition for the common resource resulting lower biomass accumulation (Abuzar*et.al.,* 2011; Adeland Soleymani, 2015).

Thousand seed weight

The result of this study showed significant variation on thousand seed weight. The highest thousand seed weight was recroded at 71,428 plants ha^{-1} (70cm x 20cm) followed by 83,333plants ha^{-1} (60cm x 20cm) but statistically at par (Figure 5).

It is obvious that seed size has an implication on germination capacity and seedling vigor as different seed size have various food accumulation.

Other research findings also revealed that various agronomic factors significantly influence size of maize seeds, which in turn influence the germination capacity and seed vigor as it depends on the accumulation of starch (Sulewska*et al.,* 2014).

The same authors described that one of the factors affecting seed vigor is the activity of amylases catalyzing hydrolytic degradation of starch into simple sugar, which are substrates necessary for metabolic processes, conditioning the growth and development of the embryo. This implies that appropriate plant spacing for basic seed production is very crucial to harvest optimum seed yield with better seed size to ensure higher germination capacity and seed vigor capacity. (Sulewska*et al.*, 2014; Kandil *et al.,* 2017).

Germination capacity

The result of the study indicated that the highest germination capacity (97%) of the seed was recorded at 60 cm and 75 cm inter row spacing and same intra row spacing (20 cm) while the lowest germination capacity

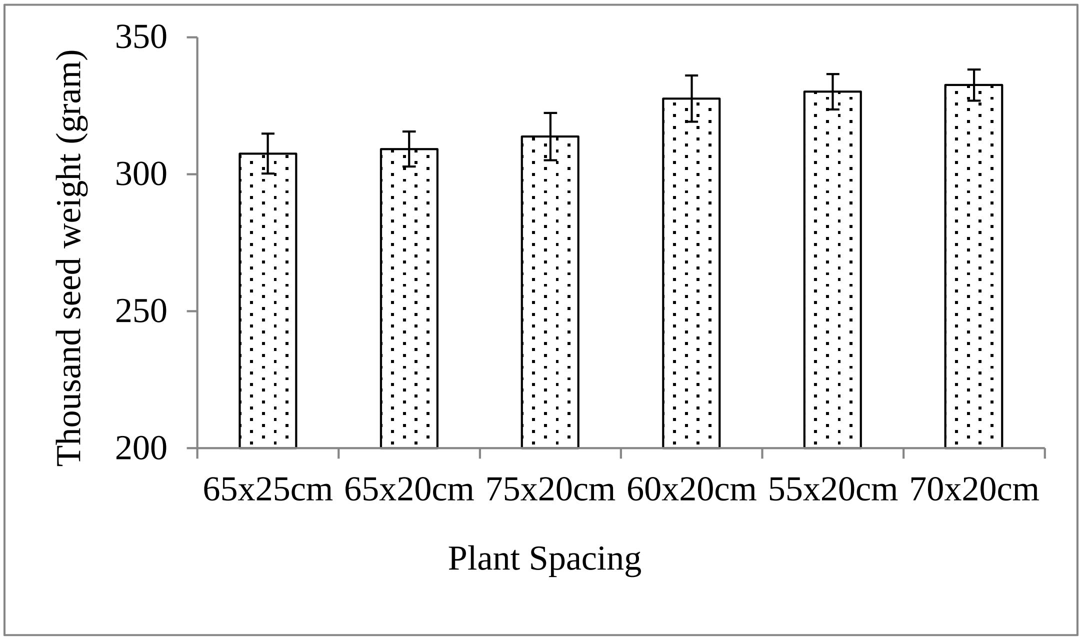

Figure 5: The effect of plant population on thousand seed weight basic seed of female Maize at Bako, 2017-2018

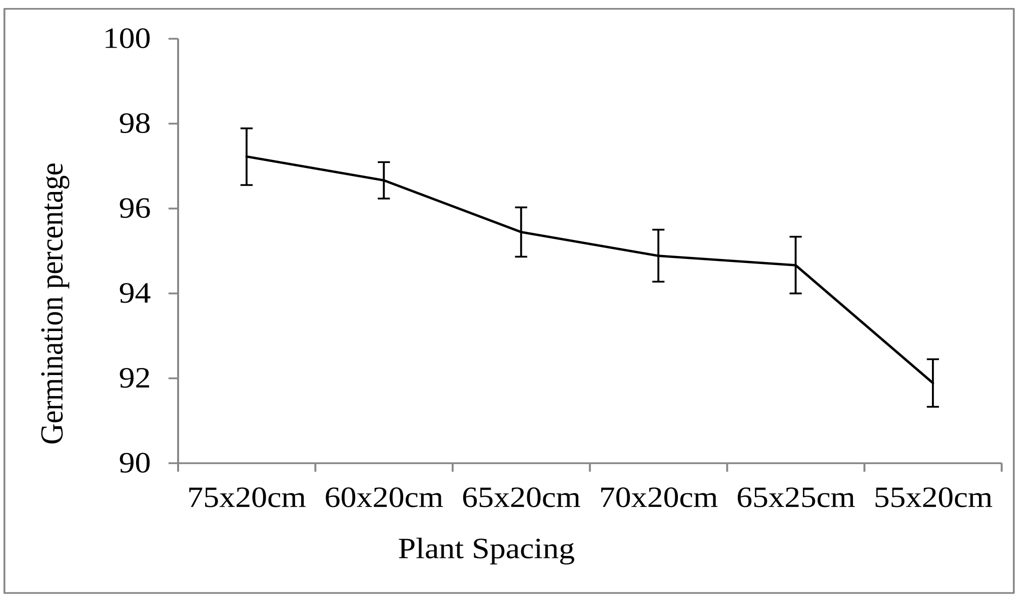

Figure 6: The effect of plant population on germination capacity of basic seed of female maize at Bako, 2017-2018

(92%) was recorded at the highest plant population with 55cm inter and 20 cm intra row spacing(Figure 6).

Even though the germination percentage under all various plant population fit the Ethiopian standard (ESA, 2012) made for spacification for hybrid seed (minimum germination capacity for basic seed production is 85%), a significant variation of germination capacity has an implication on seed rates to get optimum plant population that directly impact on the productivity. For instance, basic seed having 97% germaination capacity has relatively lower seed rates than seeds having 90% of germaniation capcity.

Economic benefit

Even though yield gained due to application of 111 $kgha^{-1}$ and 137 $kgha^{-1}$ of N was statistically at par, more than 49% (4366 ETB ha^{-1}) of extra net absolute profit could be obtained when maximum N rate was used as compared to yield gained under application of 111 kg ha^{-1}(Table 3).

Table 3: Adjusted absolute yield response (AAYR), relative yield response (RYR), absolute input cost (AIC), absolute marginal profit (AMP) and absolute net benefit (ANB) under different treatment levels of nitrogen rate and plant population against the standard checks at Bako, 2017-2018

Nitrogen rate (kg ha^{-1})	AAYR(kg ha^{-1})	RYR (%)	AIC (ETB ha^{-1})	AMP(ETBha^{-1})	ANB (ETBha^{-1})
111	218	7%	986	9801	8815
137	337	10%	1988	15169	13181
88*	0	0	0	0	0
Plant population					
65x25cm	-290.02	-9%	-265	-14501	-14236
70x20cm	-86.82	-3%	234	-4341	-4575
65x20cm	-73.80	-2%	510	-3690	-4200
55x20cm	302.32	10%	1215	15116	13901
60x20cm	485.70	16%	833	24285	23452
75x20cm**	0	0	0	0	0

Note that *= current N application rate for maize basic seed production as standard check and **= current use of plant population as standard check; AIC=Additional input cost (calculated as extra input cost due to various treatment levels minus input cost of the standard check multiplied unit price (52.80 ETB kg-1 of inbred line seeds, 13.54 ETB kg^{-1}fertilizer in the form of urea and 75 ETB per person of daily labor); AMP= absolute marginal profit (calculated as adjusted basic seed yield obtained under different treatments minus yield obtained under standard checks multiplied by 50 ETB kg^{-1});ANB= Absolute net Benefit (calculated as absolute marginal benefit minus absolute input cost

This may related with relatively higher basic seed price that makes profitable under intensive application of N fertilizer compared to certified hybrid maize seed. Of course, both 111 kg ha^{-1} and 137 kg ha^{-1}application significantly recorded 13181 ETB ha^{-1} and 8815 ETB ha^{-1} net absolute profit over the standard check (Table 3). This implies that intensive fertilization for commercial investor is preferable to enhance the profitability per unit area whereas use of 111 kgha^{-1} is very much better than the previous recommendation, and it is advisable for small scale seed business.

On other hand, positive adjusted absolute responses were recorded only under the uses of two consecutive maximum plant populations while other remaining plant population showed yield penalty compared to the standard check (66,666 plant ha^{-1}). While positive absolute yield gain over the standard check was 7% at 90,909 plants ha^{-1} and 10% at 83,333 plant ha^{-1} (Table 3), yield reduction was varied from 2% at 76,923 plants ha^{-1} to 9% at the lowest plant population as compared against the standard check, and these practices showed negative profit indicating that standard check is preferable. Even though yield obtained under practice of 83,333 plants ha^{-1} (60cmx20 cm) and 90,909 plants ha^{-1} showed statistically at par, lesser plant population had lower absolute input cost and higher absolute net benefit. In other words, practicing 60 cm inter and 20 cm intra row spacing having 83,333 plants ha^{-1} population earn 9551 ETB ha^{-1} surplus of net profit when compared with net profit gained under the highest plant population, and it's the best practice to boost productivity and profitability for production of hybrid maize basic seeds.

CONCLUSION AND RECOMMENDATION

Optimum agronomic practice is one of the best choices to enhance the productivity of hybrid maize female basic seed production. The previous recommendation of plant spacing for planting inbred lines was relatively wider as the plant canopy and vigor capacity of the inbred lines are relatively very low. The result of this study revealed that main effects of nitrogen rate and plant population showed significant effects on plant height, yield, biomass and thousand seed weight. The highest significant yield increase was recorded under practice of 83,333plants ha^{-1} with 60cm inter and 20 cm followed by 90,909 plants ha^{-1} (55cm x 20cm), and the relative yield increase over the standard check is 16% and 10%, respectively. On the other hand, the yield of female basic seed was significantly increased by 7% and 10% when 111 kg Nha^{-1} and 137 kg Nha^{-1} of nitrogen rate were, respectively, applied as compared to the standard check (88 kg Nha^{-1}). The highest positive absolute economic net benefit was also obtained by practicing 83,333plants ha^{-1} plant population, which is the best practice to enhance productivity and profitability. On the other hand, the highest absolute net benefit was recorded under the use of 137 kg Nha^{-1} followed by 111 kg Nha^{-1} over the standard check.

As of recommendation, practice of 83,333 plants ha^{-1} of plant population by practicing 60 cm inter and 20 cm intra row spacing a seed rate of 20.7 kg ha^{-1}(16 kg ha^{-1} of CML-395 and 4.7 kg ha^{-1} of CML-202) with at least 90% germination capacity is recommended for seed producers.

On the other hand, intensive fertilization (137 kg ha^{-1}) for commercial investor is preferable to enhance the profitability per unit area whereas use of 111 kg ha^{-1} is very much better than the previous recommendation, and it is advisable for small scale seed business.

ACKNOWLEDGMENT

The authors greatly acknowledge Oromia Agricultural Research Institute for financial support. The authors also sincerely acknowledge Bako Agricultural Research Center for all technical and managerial supports and great respects also go to the seed technology and multiplication unit of the center for successful execution of the experiment.

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