Vol. 12(8), pp. 155-163, August 2018 DOI: 10.5897/AJPS2017.1557 Article Number: 026664958316 ISSN 1996-0824 Copyright © 2018 Author(s) retain the copyright of this article http://www.academicjournals.org/AJPS



African Journal of Plant Science

Full Length Research Paper

Evaluation of faba bean (*Vicia faba* L.) varieties for yield and reaction to chocolate spot disease at Chencha, Southern Ethiopia

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Received 6 April, 2017; Accepted 12 May, 2017

Field experiment was conducted at Chencha Woreda with the objective of evaluating faba bean varieties for better yield and chocolate spot disease resistance during 2015 main cropping season. Ten improved faba bean varieties and two local controls were evaluated in randomized complete block design (RCBD) with three replications. Field performances of varieties were evaluated for grain yield and yield parameters on sampled plant. Chocolate spot disease severity was recorded following 1 to 9 disease scale and used for area under disease progress curve (AUDPC) calculation. There was significant (P < 0.05) difference among the varieties for yield and resistance to chocolate spot disease. Higher yield was recorded in *Moti, Gora* and *Wolki* varieties while low yield was recorded from Hachalu and Degaga. Maximum chocolate spot severity and AUDPC were recorded on *Baela, Degaga, Gora* and *Gebelcho* varieties while minimum disease severity of 29.63, 24.69, 25.93 and 25.93%, respectively. *Moti, CS-20DK*, *Hachalu* and *Tumsa* varieties showed moderately resistant to chocolate spot disease severity while *Dosha* was moderately susceptible varieties. From the result it could be concluded and recommended that varieties *Moti, Gora* and *Tumsa* are promising at high lands of Chencha, and may used them in larger plot for identification of adaptable variety.

Key words: Faba bean, varieties, yield, disease severity, chocolate spot.

INTRODUCTION

Faba bean (*Vicia faba* L. 2n = 12), is a legume member belonging to the family Fabaceae. Species in genus *vicia* are genetically separated from each other according to differences in some of the seed characters such as weight, shape and size (Hawtin and Hebblethwaite, 1983). Genetic variability of faba bean is quite large. The great variability may be due to the presence of intermediate crossing system between autogamy and

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Author(s) agree that this article remain permanently open access under the terms of the <u>Creative Commons Attribution</u> <u>License 4.0 International License</u> allogamy (Hanelt and Mettin, 1989). In fact, *V. faba* is partially pollinated by insects, so the pollinators can carry out both self-pollinations by the tripping process when they trip the flower and out crossing and when they visit other plants flowers (Nadal et al., 2003).

Crop diversification and low input agriculture are major drivers for sustainable agricultural policy. Pulse crops can contribute positively to these two policy goals because, as a result of biological nitrogen fixation, they require minimal inputs and provide a very effective break crop in a cereal dominated rotation. Pulse crops are also an efficient source of plant-derived protein for food and animal feed. In Ethiopia, three pulse crops (faba bean, field pea and fenugreek) occupied an area of 759,782.79 ha with a grain yield of 8.74 million quintals and its productivity is 10 to 45 quintal/ha across the country (CSA, 2013a).

Ethiopia is one of the largest producer of faba bean in the world second only to China (Hawtin and Hebblethwaite, 1983). The country is considered as the secondary center of diversity and also one of the nine major agro-geographical production regions of faba bean (Telaye et al., 1994). The production is mainly concentrated in the high-altitudes of Ethiopia ranging between altitudes 1800 and 3000 m.a.s.l with annual rainfall ranging from 700 to 1100 mm (Telaye, 1988). The study area has a total area of 37,360 ha land from this faba bean and field pea shares 2731 and 2285 ha of land annually, from these crops; faba bean ranks first in pulse crops and considered as the most important pulse crop in the area (Sankura. 2015 personal communication). Faba bean serves as a daily food and as cash crop in many parts of the country (Hawtin and Hebblethwaite, 1983). The pulses production and productivity is constrained by several biotic and abiotic stresses, of which lack of improved varieties, shortage of certified seeds, diseases such as rust, powdery mildew and root rot, insect pests such as aphids and low soil fertility are the major ones and becoming a major challenge to food security. In addition to this, its production in Ethiopia is limited and fails to face the increasing local consumption of seeds due to gradual decreases in its average yield. So, increasing crop production is the major target of the national agriculture policy and can be achieved by growing high yielding and stable cultivars under favorable environmental conditions (Graham and Vance, 2003).

This can be achieved by continuous highland pulse research to develop high yielding, pest resistance/tolerant, excellent in other agronomic traits, high quality, and widely adapted varieties that suit different cropping systems and farming conditions. Moreover, adequate seeds of released varieties should be made available to small scale farmers and commercial producers. The principal aim of variety trial research on highland pulses is generally to contribute to the general development policy of the nation by increasing production, productivity and thereby increasing income-generation capacity for the farmer through testing high-yielding varieties with stable performance and disease resistance. Agricultural office in the study area mentioned many problems in their extension system with the farmer associated with the production and productivity of faba bean. Among them lack of improved varieties, introduction of new varieties to the locality without conducting variety trial by only considering similar agro-ecological conditions and there are also insect pests affecting with the growth performance of this crop.

Besides these, the production of Faba bean is insufficient as a result of low crop yields because farmers grow varieties that are susceptible to diseases, insect pests, drought and high summer temperatures (ICARDA, 2008). FAOSTAT (2008) report showed that Faba bean production has declined from 4.8 million ha in 1961 to 2.4 in 2008 with 4.8 to 4.4 tons per hectare reduction in production. This reduction was due to susceptibility of faba bean varieties to biotic factors (Sillero et al., 2010) and abiotic stresses (Link et al., 2010). Chocolate spot (*Botrytis fabae*), Ascochyta blight (*Ascochyta fabae*) and faba bean rust (*Uromyces viciae-fabae*) are identified as the major diseases affecting faba bean in the country (ICARDA, 2008). In fact, chocolate spot and rust became the major threat worldwide in faba bean production.

In Ethiopia, the productivity of faba bean is far below its potential due to the aforementioned factors (Mussa et al., 2008). Winch (2006) reported that, the productivity of faba bean in Ethiopia is guite lower (15.2 gt/ha) (CSA, 2013b), as compared to that in UK, which is about 30 gt/ha). In Ethiopia, there are about 29 improved faba bean varieties which are adapted to different agroecology with varying reaction to diseases (Crop variety register issue No.17, 2014). Farmers in the Ethiopia commonly used to cultivate local varieties (Thijssen et al., 2008). In most cases local varieties are expected to be low yielding and susceptible to both biotic and abiotic stresses. Local landraces faba beans are highly susceptible to the disease and give low yield (Samuel et al., 2008). Chocolate spot disease is among the major diseases of faba bean which becomes the major threat in the study area. Therefore, growing of high yielding and disease resistance varieties of faba bean is crucial to ensure the sustainability of the crop and food security. Thus the experiment was conducted to evaluate improved faba bean varieties and local check for screening high yielding variety and assessing reaction to chocolate spot disease resistance in field condition to boost the productivity of the crop.

To intervene the aforementioned problems, this study was initiated with the general objectives specific in the highland pulse crops research strategy, the specific objectives to select and promote high yielding and well adapted faba bean varieties with desirable agronomic and quality traits for the highlands of Gamo Gofa area specifically in Chencha Woreda.

Objectives

The general objective is to evaluate faba bean genotypes for better yield and resistance to chocolate spot disease. The specific objectives include the following:

1. To identify promising faba bean varieties for seed yield and other important agronomic traits.

2. To assess the reaction of faba bean varieties to chocolate spot disease

3. To recommend promising varieties for further study in those important traits

MATERIALS AND METHODS

Experimental area description

The study was conducted at Chencha Woreda, Arba Minch University Gircha research Center, Chencha town. Chencha is found in the Gamo Gofa administrative zone of the Southrn Nation Nationalities People Republic of Ethiopia. The altitude is ranging between 1600 and 3200 m a.s.l. The mean annual temperature and rainfall of the study areas are 22.5°C and 1100 to 1600 mm/annum, respectively. The altitude of Gircha research is ranges up to 3007 m.a.s.l. The soil condition is characterized as 60, 30, 6 and 4% brown, black, grey and black in color, respectively and the topography of the Woreda is described as 65, 17, 13 and 3% mountainous, steeply, valley and flat, respectively (Sankura, 2015, personal communication). According to our observation the soil texturally looks like clay loam with brown color.

Plant material and planting

Faba bean cultivars namely Degaga (R-878-3), Gora (EK01024-1-2), Dosha (COLL 155/00-3), Gebelcho (EH96006-1), Hachalu (EH00102-4-1), Wolki (EH96049-2), Moti (EH95078-6), CS-20DK, Tumsa (EH99051-3), and Obse (EH95073-1) and local checks namely Baela and Orde Baela were used in this trial. The seeds of the cultivars were collected from legumes research coordinating centers (Kulumsa and Holeta Agricultural research centers) in Ethiopia. Recommended fertilizer rate was added during seed planting. Even though, the crop is nitrogen fixer in nature, recommended dose of nitrogen source fertilizer (Urea) was added to initiate nodulation process. Planting was done using seeds of faba bean at recommended rate per kilogram of seeds, in hills (two seeds/hill) to guarantee the germination of the seeds on the two sides of the ridges in row. Thinning was practiced after 21 days from sowing to secure one plants/hill. All the other recommended cultural practices for growing faba bean were followed like weed control, insect control, watering and others.

Experimental design

Ten faba bean cultivars advanced from pre varietal trial was tested along with two standard checks (local varieties) were planted on cropping season of June 2015 to December, 2015. Randomized Complete Block Design (RCBD) with three replications was used. The plots consisted of five rows of 4m length with inter- and intrarow spacing of 15 and 60 cm, respectively. Plot size was arranged as 4 m (length) × 2 m (width). The total area for this experiment was around 907.2 m². The experimental field was managed as per the standard field plot techniques and standard agronomic practice in the season.

Data collection

Plant heights (cm) at flowering at different interval were measured on 5 representative sample plants of faba bean cultivars. At harvesting time, field performance evaluations like seed yield (kg/m²), 1000 seed weight in gram, number of pod per plant, number of seed per pod, The disease severity of chocolate spot was recorded from 65 to 86 days from sowing in 7 day intervals for 3 times to see disease severity at different growth stage using the scale of Bernier et al. (1993), where, 1 = no disease symptoms or very small specks (highly resistant), 3 = few small disease lesions (resistant), 5 = some coalesced lesions, with some defoliation (moderately resistant), 7 = large coalesced sporulating lesions, 50% defoliation some dead plants (susceptible), 9 = extensive, heavy spourlation, stem girdling, blackening and death of more than 80% of plants (heavily susceptible). The response of tested varieties were classified into six reaction groups according to Abo-Hegazy et al. (2012), where 0 to 2% is highly resistant (HR), > 2 to 15% is resistant (R), > 15 to 40% is moderately resistant (MR), > 40 to 60% is moderately susceptible (MS) and > 60 to 80% is susceptible (S) >80 to 100% is highly susceptible (HS) based on percent disease severity values.

Percent severity index (PSI%) was calculated using:

$$\mathsf{PSI}(\%) = \frac{\sum(\mathsf{NPC^*CR})^*100}{\mathsf{NIP^*MSC}}$$

Where NPC = number of plants in each class rate, CR = class rate, NIP = number of infected plants and MSC = maximum severity class rate.

Area under disease progress curve (AUDPC) was calculated using the formula adapted from Cooke (2006) as followed by using the disease severity score of each plot in fixed interval of 7 day.

AUDPC =
$$\sum_{i=1}^{n-1} [0.5 (yi + 1 + yi)(ti + 1 - ti)]$$

Where y_i = the average coefficient of infection of the ith observation, y_{i+1} = the average coefficient of infection of the i+1th observation and; t_{i+1} - t_i = the number of days between the ith observation and i+1th observation, and n = number of observations.

Data analysis

Analysis of variance (ANOVA) was conducted to see cultivar differences from the mean values of the sampled plant based on the experimental design used by statistical software (Gene Stat. 15th edition). Fisher's LSD test at 5% level of significance was performed to determine whether there were significant differences among the cultivars for all measured traits. The coefficient of variation was calculated based on Burton (1952). Broad sense heritability ($h^2 = S^2g/S^2p$ or $h^2 = S^2g/S^2g + S^2e$) where, $h^2 =$ heritability, S^2g = variance due to genotype, S^2p = variance due to phenotype = the sum of variance due to genotype and the experimental variance) was calculated according to (Johnson et al., 1955). The Pearson correlation coefficient between all measured trait means for twelve cultivars was determined.

RESULTS AND DISCUSSION

Growth and yield related traits

Faba bean varieties were significantly different (P < 0.05) for plant height (Figure 1A). The tallest plant heights were

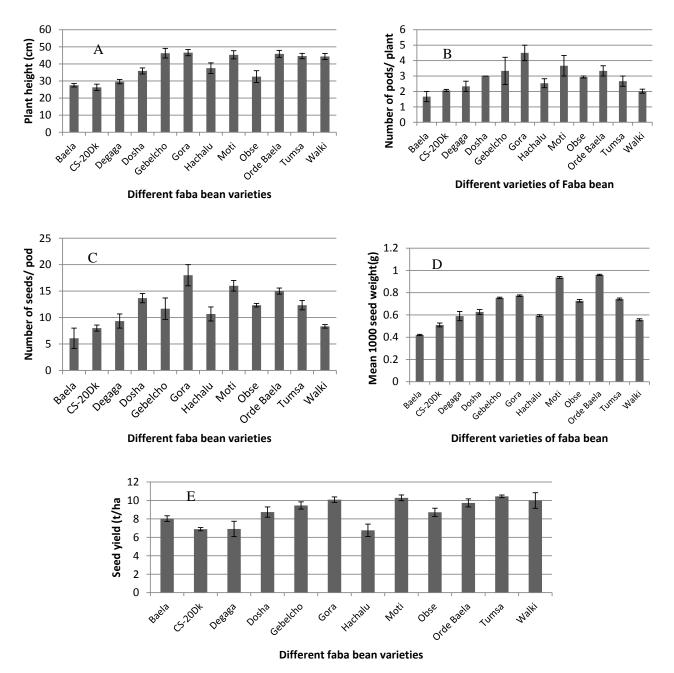


Figure 1. Effect of varieties on yield and yield components (A-E) of 10 improved and 2 local faba bean varieties during the main season of 2015/16 at Chencha Gircha research station Arba Minch, Ethiopia. Varieties were significantly (P < 0.05) different for all measured traits. Error bars are drawn from standard error of the mean.

recorded in Gora and Gebelcho with 46.6 and 46.27cm tall, respectively. This result disagreed with the result of Ashenafi and Mekuria (2015). They reported that Gebelcho had the shortest plant height at Sinana and Agarfa site, Ethiopia. This could be related to altitude difference of the study site. In most cases, the area becomes cooler as increasing altitude and plant metabolic activity becomes slower. On the other hand, the shortest plant heights were recorded in CS-20DK and local Baela varieties with a height of 26.3 and 27.53 cm,

respectively. Thus, CS-20DK and local Baela varieties can be considered as dwarf varieties. Talal and Munqez (2013) reported that plant height differ significantly in faba bean accessions. Faba bean genotypes showed significantly different plant height of under rain fed conditions (Della, 1988).

Number of dry pods per plant was significantly different (P < 0.05) among all the tested varieties (Figure 1 B). Gora, Moti and Gebelcho ranked first to 3rd for number of pod per plant (4.5, 3.67 and 3.33). The result for

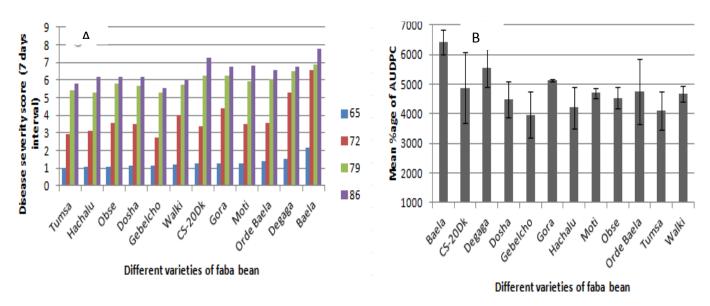


Figure 2. Disease severity score (A) from 65-86 DAS and the area under disease progress curve (AUDPC) (B) values on 10 improved and 2 local faba bean varieties 2015/2016 cropping season. Error bars are drawn from standard error of the mean.

Gebelcho was in line with the work of Ashenafi and Mekuria (2015) who reported that this variety had a higher number of pods per plant followed by variety Degaga at Agarfa experimental site. However, in the study of Tafere et al. (2012), Gebelcho and Moti varieties had the smallest number of pods per plant, which disagreed with our result. Whereas, Wolki from improved varieties and Baela from local check produced the lowest number of pods/plant. Faba bean genotypes significantly varied in production of pods per plant (Hassan and Ishaq, 1972; Pilbeam et al., 1992).

The analysis of variance revealed that there were significant (P < 0.05) differences between faba bean varieties in yield of dry seed (Figure 1E). The total grain yield recorded in the study was lower than the national average yield of faba bean; this could be due to strong acidic nature of the soil in the study area. In this study, Moti (1.028 t/ha) and Gora (1.008 t/ha) produced dry seed yield, which is the highest yield recorded. However, Degaga and CS-20DK produced lowest yield with 0.691 and 0.6887 t/ha, respectively. Following Gora and Moti varieties higher yield was obtained from Wolki, Orde baela and Gebelcho varieties at the study area with average seed yield of 0.999, 0.973 and 0.945 t/ha, respectively. Seed yield obtained from Wolki was in line with the result reported by Ashenafi and Mekuria (2015) at Sinana district even if there was differences the amount of yield obtained per hectare. However, Moti produced lowest grain yield in their study at this district. The report of ICARDA (2008) showed that shifting from traditional varieties to improved ones can bring an increment in yield of 18% in Egypt, 8% in Sudan and 42% in Ethiopia as reviewed by Ashenafi and Mekuria (2015). In this study, 30% increment of yield was found for using *Moti* variety (high yielder) as compared to Baela local variety (low yielder) at the study area.

There were significant variations in the values of 1000 seed weight shown by the faba bean varieties, confirming that the genetic variations among varieties. Of all tested varieties Orde Baela (969 g) produced heaviest seed weight followed by Moti (093 g), Gora (77 g) and Gebelcho (75 g) while, Baela resulted the lowest 1000 seed weight followed by CS-20DK, Wolki, Hachalu and Degaga (Figure 1D). This result indicated that Orde Baela local, Moti, Gora and Gebelcho varieties can be considered as large seeded while Baela local, CS-20DK, Wolki, Hachalu, and Degaga are small seeded. Seed size is one of the most important characters used for farmer to select variety. Hachalu was small seeded variety in this study which resulted in reduced seed yield. The result was in line with the work of Ashenafi and Mekuria (2015); they reported that Moti and Gebelcho varieties had the higher 100 grain weight while Degaga variety had the lowest grain weight. This result also coincides with the result of Tamane et al. (2014). The varieties evaluated in this study showed significant (P < 0.05) in number of seeds per pod. Gora had more number of seeds per pod followed by Moti. This had significant positive correlation with grain yield of faba bean varieties.

Disease severity

The intensities of disease occurrence varied at different intervals. Chocolate spot disease became sever as increasing time from the start of severity score (Figure 2). The result showed that there was a significant difference (P < 0.05) in disease severity score at 65 DAS among

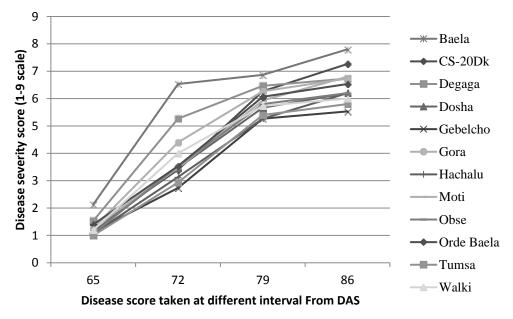


Figure 3. Disease Progress curve (DPC) for chocolate spot disease of faba bean varieties at Gircha research site Arba Minch, Ethiopia with different levels of resistance during the mainseason of 2015/16.

faba bean varieties. However, percent disease severity was not significantly different (P > 0.05). In our experimental location the lowest disease severity was recorded in variety *Tumsa, Moti, Hachalu* and *CS-20DK* varieties, that is, 29.63, 24.69, 25.93 and 25.93%, respectively, while maximum disease severity was recorded in *Degaga, Gebelcho, Dosha* and *Orde Baela* varieties, that was, 32.72, 33.95, 41.98 and 33.95% varieties at experimental site.

The result of this experiment indicated that the reaction of the individual varieties of faba bean for chocolate spot disease was more or less similar with the result of Mekuria and Ashenafi (2015). They reported that Tumsa as a resistant to chocolate spot disease. Similarly, Terefe et al (2012) and Tamene et al. (2015) reported that Tumsa variety was resistant to chocolate spot and Gebelcho variety was moderately resistant. The result was not inline to with the work of Mekuria and Ashenafi (2014). They reported that *Moti* and *Hachalu* had a moderate chocolate spot severity. This variation could be due to the difference in environmental conditions as the occurrence of disease chiefly depends on environmental condition.

There were Woreda differences among the faba bean varieties in the disease progression in the study area where the severity levels were more pronounced on all plants of the cultivar; *Dosha*, *Wolki* and *Degaga* during all the successive assessments 65 DAS as opposed to that of *Tumsa*, *Hachalu* and *Moti* (Figure 3). In general, the final chocolate spot severity was not significantly different (P > 0.05) among the varieties (Table 1). The highest chocolate spot infections on the local cultivars were

33.9%, as opposed to the lowest mean severity of 24.69% on Moti and 25.93% CS-20DK at the study site (3000 m a.s.l) (Table 1). Gora and Tumsa varieties had relatively lower infection values of 29.63 and 29.63% similar to the local check namely Baela. Gebelcho, Obse and Degaga had moderate infections that varied from 33.95, 32.41 and 32.72%, respectively. Accordingly, the four faba bean varieties namely CS-20DK, Degaga, Moti, Tumsa and Hachalu expressed relatively moderately resistance to chocolate spot at the study area natural condition. Thus, the uses of moderately resistant cultivars are recommended instead of depending solely on fungicides (Bouhassan et al., 2004; Josefina et al., 2010). In general, moderate resistance to chocolate spot with partial dominance has been reported in different researches obtained in this study.

Heritabilities of the tested varieties were found to be higher in all measured traits (Table 2). The heritabilities of most traits were above 0.5, indicating that more than 50% of the phenotypic variation observed for these traits was attributed to genetic factors. However, percent disease severity, and AUDPC showed low heritability. This could be due to severity of disease is mainly facilitated by environment.

Correlation between studied traits

Positive significant correlations were found between seed yield and plant height, number of pod per plant, number of seed per pod and 1000 seed weight (Table 3). The result was in agreement with (Silim and Saxena, 1992).

Variety	Disease severity percent	Reaction group		
Baela	29.63	MR		
CS-20Dk	25.93	MR		
Degaga	32.72	MR		
Dosha	41.98	MS		
Gebelcho	33.95	MR		
Gora	29.63	MR		
Hachalu	25.93	MR		
Moti	24.69	MR		
Obse	32.41	MR		
Orde Baela	33.95	MR		
Tumsa	29.63	MR		
Wolki	33.95	MR		
S.e.d	7			
LSD (?%)	14.5			
CV (%)	27.5			
P-value	0.539			

Table 1. Chocolate spot severity percent (%) on 10 improved and 2 local faba bean varieties at Chench Gircha research station, Arba Minch, Ethiopia during 2015/2016 main crop season.

 a DSI = mean disease severity percent assessed at 86 days after sowing (DAS). Means are not significantly different (P > 0.05) based on LSD value Test. Faba bean varieties with values >15 - 40 are considered moderately resistant while values >40 - 60 moderately susceptible (Abo-Hegazy et al., 2012).

Table 2. Heritabilities of different traits measured on 10 improvedand 2 local faba bean varieties at Chench Gircha research station,Arba Minch, Ethiopia during 2015/2016 main crop season.

S/N	Measured traits	Heritability (%)		
1	Mean of plant ht	85		
2	Number of pod/plt	65		
3	Number of seed/pod	81		
4	1000 seed wt.	97		
5	Yield (qt/ha	78		
6	disease severity score	50		
7	% disease severity	30		
8	AUDPC%	32		

Table 3. Correlation analysis among plant height, pod per plant, seed per pod, 1000 dry seed weight AUDPC, diseases severity score, percent diseases severity and grain yield of 10 improved and 2 local faba bean varieties at Chencha Gircha research station, Arba Minch, Ethiopia during 2015/2016 main crop season.

Parameter	Plant height (cm)	Pod/pla nt	Seed/pod	Yield (Kg/ha)	1000seed weight	AUDPC (%)	Disease severity score	Disease severity (%)
Plant height (cm)	-							
pod/plt	0.62*	-						
seed/pod	0.65*	0.90***	-					
yield (Kg/ha)	0.69**	0.45*	0.53*	-				
1000seed weight	0.68**	0.65*	0.7719**	0.58*	-			
AUDPC (%)	-0.49 ^{ns}	-0.44*	-0.44*	-0.27 ^{ns}	-0.34 ^{ns}	-		
Disease severity (score)	-0.30 ^{ns}	-0.17 ^{ns}	-0.254 ^{ns}	-0.04 ^{ns}	-0.25 ^{ns}	0.37 ^{ns}	-	
Dss sev (%)	0.07 ^{ns}	0.08 ^{ns}	0.20 ^{ns}	0.15 ^{ns}	-0.03 ^{ns}	0.06 ^{ns}	0.04 ^{ns}	-

* $P \le 0.05$; ** $P \le 0.01$; *** $P \le 0.001$; ns = non-significant.

They reported that there is a positive relationship between seed yield and hundred seed weight, biological yield and seed per pod. Pod per plant, 1000 seed weight and number of seed per pod were positively significantly correlated with each other. These results suggested that for a high seed yield, fewer pods with large seed might be compensating for the low pod number per plant. The positive correlations observed between seed yield and 1000 seed weight might indicate the significance of the seed size in determining the final yield of faba bean varieties. Negative non-significant correlation between seed yield and AUDPC, disease severity score, and percent disease severity was recorded in this study. This indicates disease plays a vital role in reduction of yield.

CONCLUSION AND RECOMMENDATION

Ten improved and 2 local check faba bean varieties were evaluated for their yield, yield components and reaction to chocolate spot disease at Chencha Gircha research center Arba Minch, Ethiopia. There were variations between the varieties for most of the traits measured. The higher seed yield was recorded in variety Moti which was followed by variety Gora, Wolki and Gebelcho whereas varieties Hachalu and CS-20DK produced lower seed yield. Therefore, varieties Moti, Gora and Wolki are promising for grain yield at Chencha area. The varieties tested in this study produced lower yield compared to the national average yield of faba bean in Ethiopia, this could be associated with strong acidic characteristics of the soil. Regarding to faba bean varieties response to chocolate spot diseases under natural condition. It can be concluded from the current results that those faba bean varieties Moti, CS-20DK, Gora and Tumsa showed moderate resistance to B. fabae infection under field condition and superior yield especially in Moti and Gora are recommended to be adapted in chocolate spot prone areas of the study site and adjacent areas, of south Ethiopia. In addition, except variety Dosha, which is moderately susceptible, can also be grown in areas with contrasting environments within the faba bean production area. Using improved variety had an advantage of more than 30% increment of yield compared to Baela local variety at the study area. The varieties tested in this study produced lower yield compared to the national average vield of faba bean, this could be associated with strong acidic characteristics of the soil. This problem would lead to inefficient exploitation crops inherent capacity to use nutrient and produce potential yield. The promising varieties identified in the study may be further used at the same and other locations for confirmation of results on the performance of faba bean variety.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

AKNOWLEDGEMENT

The authors want to thank Arba Minch University Research Directorate office for their financial support and Chencha woreda Agricultural office and for their active participation when the experiment was conducted.

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