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Genetic Variation and Gains from Selection for Symbio-Agronomic Performance in Ethiopian Chickpea (*Cicer arietinum* L.) Germplasm Accessions

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ABSTRACT

Information on traits relationship, genetic variation and gains from selection for symbiotic and agronomic characters in chickpea (*Cicer arietinum* L.) are limited. An experiment was undertaken at two locations (Ginchi and Ambo) in Ethiopia in 2009/2010 to assess the relationship, genetic variation and genetic gain from selection for attributes of symbiotic and agronomic significance. The difference technique with genetically non-nodulating chickpea reference was employed to estimate the amount of symbiotic nitrogen fixation. Significant positive correlations were found between a number of symbiotic and agronomic traits. Grain yield was positively associated with fixed nitrogen assimilation efficiency (r = 0.39), shoot (r = 0.31), grain (r = 0.93), and above ground biomass nitrogen yields (r = 0.77) and NHI (r = 0.52). Grain yield was also positively influenced by agronomic characters including grain filling period (r = 0.38), pod (r = 0.57) and seed numbers (r = 0.59), shoot (r = 0.67), and total above ground biomass (r = 0.79) accumulations, HI (r = 0.53), grain production efficiency (r = 0.92) and biomass production (r = 0.81) and economic growth (r = 0.93) rates. Characters like shoot, grain and total biomass nitrogen contents and fixation, fixed nitrogen assimilation efficiency, seed size, grain filling period showed higher genetic variation, broad-sense heritability and expected genetic gains from selection. The Z-test revealed effective selection at phenotypic level for all traits. Further implications of the study in terms of selection strategy have also been discussed.

Keywords: broad-sense heritability, correlation coefficient, genetic gain from selection, Z-test

INTRODUCTION

Chickpea (*Cicer arietinum* L.) is known to be grown in Ethiopia since antiquity (Muehlbauer and Tullu 1997; Bejiga and van der Maesen 2006). Some sources indicate that the crop was grown in Ethiopia as early as 1520 BC (Joshi *et al*. 2001). Despite the ancient history of the crop and its economic and ecological significances in fixing atmospheric nitrogen and improving soil fertility, the productivity of chickpea in Ethiopia is below the attainable genetic potential of the crop (Muehlbauer and Tullu 1997) and the productivity in many other countries (Bejiga and van der Maesen 2006; CSA 2011). This has been attributed to low productivity of the local chickpea cultivars grown in Ethiopia and poor soil fertility with little application of production inputs particularly fertilizers (Getachew *et al*. 2006).

A number of workers advised that agricultural sustainability should rely on the use and effective management of internal resources like integration of cereals with legumes (Bohlool *et al*. 1992; Pearson *et al*. 1995; Srinivasarao *et al*. 2006; Beebe *et al*. 2006). Symbiotic nitrogen fixation is considered as the safest and sustainable source of nitrogen including for organic agriculture, where sustainability of farming system and provision of healthy food for human consumption are big concerns (Wolfe *et al*. 2008; IFPRI 2010). The breeding of chickpea genotypes with better symbiotic nitrogen fixation, therefore, is a suitable approach to address soil fertility problems of the majority of resource-poor farmers (Keneni *et al*. 2012).

Breeding progress depends on the magnitude of genetic variability among the source genetic materials, heritability of a given trait in a given environment and the level of selection intensity applied (Falconer 1989; Hayward and Breese 1993; Singh 2005). To this end, existence of high heritability (broad sense, narrow sense, realized) of the legume symbiotic activity is demonstrated in many cases and this indicates that plant and strain selection for symbiotic nitrogen fixation may be highly effective (Provorov and Tikhonovich 2004; Belay and Assefa 2011).

In chickpea, both nodulation efficiency and grain yield could be improved as the result of selection (Sattar *et al*. 1995; Ali *et al*. 2002). Many reviews have been written to show that effective symbiotic nitrogen fixation could be achieved from genetic improvement of the host plant and the strain as the existence of genetic variation has been demonstrated in both the chickpea host plant and the bacterial strain (Beringer *et al*. 1988; Malhotra *et al*. 2004; Winter *et al*. 2004; Belay and Assefa 2011). Crossing genotypes from different sources followed by selection, for instance, was found to be promising for improving some symbiotic characters like nodule number and dry weight in common bean (*Phaseolus vulgaris*) (Franco *et al*. 2001).

Direct selection to improve symbiotic efficiency based on the amount of fixed nitrogen may be difficult because of the polygenic nature and the associated low genetic variation and heritability (Austin 1993; Tate 2000). Under circumstances where characters are governed by poly genes, direct selection may also be less efficient. There is also no practical way that breeders can identify best nitrogen fixing individual plants from among the segregating materials or germplasm in the breeding nursery (Keneni and Imtiaz 2010). Visual selection for better yielding individuals may

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Table 1 Description of the test locations for geographical position and physico-chemical properties of the soils.

Parameter	Source of soil	
	Ambo	Ginchi
Latitude	09° 00′ N	09° 00′ N
Longitude	37° 22′ E	38° 10′ E
Altitude (m.a.s.l.)	2225	2200
Mean annual rainfall (mm)	1000	1110
% Clay	70.00	65.83
% Silt	15.00	20.42
% Sand	15.00	13.75
Organic C (%)	1.53 (low)	1.30 (low)
N (%)	0.103 (low)	0.103 (low)
C/N ratio	14.85 (high)	12.62 (high)
P (ppm*)	18.07 (high)	4.49 (low)
K (Meq/100 g soil)	2.438 (high)	2.485 (high)
Ca (Meq/100 mg soil)	59.03 (high)	39.62 (high)
Mg (Meq/100 mg soil)	11.20 (high)	9.00 (high)
Na (Meq/100 mg soil)	0.70 (high)	0.61 (high)
SO_4 S (ppm)	5.23 (optimum)	5.62 (optimum)
Fe (ppm)	27.73 (high)	51.50 (high)
pH (1:1 H_2O)	7.23 (optimum)	6.18 (optimum)
EC (µS)**	650.00 (high)	547.33 (high)

*ppm = parts per million; **µS = micro siemens

be possible for attributes of grain yield but the problem of polygenic nature still holds. It would therefore be useful to use secondary traits that can be positively associated with the primary traits such as the amount of nitrogen fixation and grain yield, genetically variable and highly heritable and easily observable in the field (Edmeades *et al*. 1997). Despite lower heritability and genetic variance of primary traits, heritability and genetic variation of some secondary traits may remain high and at the same time the traits may maintain good level of positive correlation with the amount of primary traits of interest like symbiotic nitrogen fixation and grain yield (Edmeades *et al*. 1997). This study was designed to assess genetic variation, trait interrelationship and gains from selection for attributes of symbiotic and agronomic significance in chickpea germplasm accessions collected in Ethiopia.

MATERIALS AND METHODS

Plant materials

In this study, a total of 155 chickpea genotypes were evaluated. They include 139 accessions from different regions of Ethiopia kindly provided by the Ethiopian Institute of Biodiversity Conservation (IBC), 5 improved genotypes from the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), 8 originally introduced commercial cultivars released in Ethiopia and three genetically non-nodulating reference lines received from ICRISAT and the International Center for Agricultural Research in the Dry Areas (ICARDA). The passport description of the genotypes and the map of the areas of collection for the Ethiopian accessions are presented in **Appendices 1** and **2**. All genotypes were rejuvenated during 2009 under the same conditions in Ginchi to reasonably minimize initial variation due to the differences in seed age and indigenous seed nitrogen content (Liao *et al*. 2008).

The field evaluation test environment

The experiment was conducted under field conditions at two locations (Ginchi and Ambo) in central part of Ethiopia for one year during the main cropping season of 2009/10 (September to January). The two locations are characterized by Vertisol soils (Dibabe *et al*. 2001) and are assumed to represent the major chickpea production areas in Ethiopia. Chickpea is mostly grown on Vertisol soils with residual moisture in Ethiopia. Climatic data of the two locations during the growing period were taken from Ambo and Holetta Research Centers as presented in **Figs. 1A** and **1B**. Soil samples from both locations were collected from the rhizosphere (top 20 cm) for physico-chemical characterization (**Table 1**).

Rhizobium inoculant and inoculation

An effective strain of *Rhizobium* for chickpea, CP EAL 004, originally isolated by the National Soil Laboratory from a collection of Ada'a District of East Shewa Zone, Ethiopia, was used for the study. The isolate was found to be efficient in nodulation and symbiotic nitrogen fixation in previous studies (Hailemariam and Tsige 2006). The inoculum was received at the concentration of approximately 10^9 cells g^{-1} of peat carrier. The concentration and purity of the inoculum was confirmed in the Soil Microbiology Laboratory at the Holetta Research Center immediately before planting. Seeds of all genotypes were coated with the inoculant at the rate of approximately 2 g of inoculum for 80 seeds using 40% gum Arabic as an adhesive.

Experimental design and layout of field trials

A randomized complete block design with 4 replications was used. A blanket basal application of phosphorus was made to all plots in the form of triple supper phosphate (TSP) at the recommended rate (20 g for a single row of 4 m). Sowing rate was 5 cm between plants and 40 cm between adjacent rows. Experimental plot was represented by one row 4 m long. The genotypes were assigned to plots at random within each block. Nitrogenous fertilizer was totally omitted and all other crop management and protection practices were applied uniformly to all genotypes as required.

Shoot and grain nitrogen analysis

Forty-five days after emergence (shortly before flowering), five plants from each plot were carefully dug up and their roots washed free from soils with water running over a sieve for collection of released nodules. Representative shoot samples were collected shortly before flowering and again at 90% physiological maturity. The grain samples from each plot were oven-dried to constant moisture at 70°C for 18 h. The dry samples were ground to pass through a 1-mm mesh sieve to determine nitrogen using the Kjeldahl technique (AOAC 1970) at Holetta and Debre Zeit Soil Science Research Laboratories. The amount of total nitrogen accumulated from fixation in shoots and grains of the test genotypes was estimated by the difference method (difference in nitrogen content between the nodulating test genotypes and the non-nodulating reference check) using a non-nodulating reference genotype PM 233. Protein contents of shoot and grain were determined by multiplying nitrogen percentages by the standard conversion factor of 6.25 (AOAC 1970). Based on the nitrogen contents, the following parameters were calculated:

$$\text{N fixed in shoot} = \frac{(Nsfg - Nsnfg)X\ 100}{Nsfg}$$

where Nsfg = amount of nitrogen in shoot of fixing genotype and Nsnfg = amount of nitrogen in shoot of non-fixing genotype.

$$\text{N fixed in grain} = \frac{(Ngfg - Ngnfg)X\ 100}{Ngfg}$$

where Ngfg = amount of nitrogen in grain of fixing genotype and Ngnfg = amount of nitrogen in grain of non-fixing genotype.

$$\text{N fixed in biomass} = \frac{(N\ fixed\ in\ shoot + N\ fixed\ in\ grain)X\ 100}{Nsfg + Ngfg}$$

$$\text{N assimilation efficiency} = \frac{(N\ fixed\ in\ grain)X\ 100}{Nsfg + Ngfg}$$

Grain N yield = Grain N content × grain yield
Shoot N yield = Shoot N content × shoot yield
Biomass N yield = Grain N yield + shoot N yield
Nitrogen harvest index (NHI) was estimated as:

$$\text{NHI} = \frac{\text{Grain N yield}}{\text{Biomass N yield}}$$

Data collection on symbiotic and agronomic characters

Data were collected either on plot basis or from randomly selected five plants following the recommendations of international bodies (IBPGR, ICRISAT and ICARDA 1993).

Records were taken on symbiotic characters as follows: (1) number of nodules (5 plants^{-1}), (2) nodule dry weight (mg 5 plants^{-1}), (3) nodulation index (nodule dry weight to shoot dry weight ratio), (4) shoot nitrogen and protein contents (%), (5) shoot nitrogen fixation, (6) grain nitrogen and protein contents, (7) grain nitrogen fixation (%), (8) above-ground biomass nitrogen content (%), (9) above-ground biomass nitrogen fixation (%), (10) fixed nitrogen assimilation efficiency (%), (11) shoot, grain and above-ground biomass nitrogen yields (g 5 plants^{-1}), and (12) nitrogen harvest index (NHI; the ratio of the amount of the element in the grain relative to the amount of the element in the total above-ground biomass).

Agronomic characters recorded include: (1) early vigor as shoot dry weight (g 5 plants^{-1}) before flowering, (2) shoot dry weight at maturity (g 5 plants^{-1}), (3) shoot dry weight ratios of nodulating to non-nodulating genotypes at maturity, (4) days to 50% flowering and 90% maturity, (5) grain-filling period (the number of days from 50% flowering to 90% physiological maturity), (6) numbers of pods and seeds, (7) total above-ground biomass weight (g 5 plants^{-1}), (8) harvest index (HI) (proportion of total above-ground biomass that is grain), (9) grain production efficiency (grain filling duration divided by duration of vegetative period and then multiplied by grain yield), (10) above-ground biomass production rate (above-ground biomass weight divided by days to 90% physiological maturity and then multiplied by 100), (11) economic growth rate (grain weight divided by grain fill duration and then multiplied by 100), (12) 1000-seed weight (g) and (13) grain yield (g 5 plants^{-1}).

Statistical analysis

Frequency distribution was used to reveal the magnitude and pattern of distribution of variation in selected traits. To compare selected subsets of the 5% best genotypes for each trait within the whole population, they were sorted and means were independently computed for each character. Mean performances of the 5% best selected genotypes and the base populations (considering the whole set of genotypes as base populations) were calculated and significance of the differences was determined by the Z-test as:

$$Z = \frac{\overline{X} - \mu}{\sigma/\sqrt{n}}$$

where $\overline{X}$ is mean of selected genotypes, μ is mean of the base populations, σ is the standard deviation calculated for the base populations, and n is the number of genotypes selected from the base population for better performance.

Partitioning of the total variance into components due to genotype (σ_g^2), location (σ_l^2) and genotype by location interaction (σ_{gl}^2) effects and error variance (σ_e^2) was performed from the analysis of variance by assuming various observed mean squares equal to their expected mean squares (**Table 2**) as suggested by Singh and Chaudhary (1985).

$\sigma_g^2 = [(\sigma_e^2 + R\sigma_{gl}^2 + RL\sigma_g^2) - (\sigma_e^2 + R\sigma_{gl}^2)]/RL = (MS3-MS4)/RL$

$\sigma_e^2 = MS5$

$\sigma_{gl}^2 = [(\sigma_e^2 + R\sigma_{gl}^2) - (\sigma_e^2)]/R = (MS4-MS5)/R$

where MS3 = mean square of genotypes, MS4 = mean square of genotype by location, and MS5 = mean square of error as given in **Table 2** below.

Broad-sense heritability (h^2) was calculated as:

$h^2 = \sigma_g^2 / [\sigma_g^2 + \sigma_{gl}^2/L + \sigma_e^2/RL] \times 100$

The predicted response of symbiotic and agronomic characters to selection or the expected genetic advance (GA) from selection were calculated, assuming the selection intensity of 5%, as:

Table 2 Skeleton of combined analysis of variance (ANOVA) used in calculation of components of variation for symbiotic and agronomic characters in 155 chickpea genotypes.

Source of variation	Degree of freedom	Mean square (MS)	Expected mean square (EMS)
Locations	L – 1	MS1	$\sigma_e^2 + G\sigma_r^2 + GR\sigma_l^2$
Replications/location	L(R – 1)	MS2	$\sigma_e^2 + G\sigma_r^2$
Genotypes	G – 1	MS3	$\sigma_e^2 + R\sigma_{gl}^2 + RL\sigma_g^2$
Genotype × Location	(G – 1)(L – 1)	MS4	$\sigma_e^2 + R\sigma_{gl}^2$
Error	L (G – 1)(R – 1)	MS5	σ_e^2

$GA = K.\ \sigma_P.\ h^2$

$GA \text{ as \% of mean} = \frac{GA}{\overline{X}} \times 100$

where K = the selection differential (K = 2.06 at 5% selection intensity) and σ_P = phenotypic standard deviation (Singh and Chaudhary 1985).

Correlation coefficients between characters were estimated based on the standard procedure as:

$r = Cov_{(xy)} / sqrt\ [\sigma_x^2 + \sigma_y^2]$

where $Cov_{(xy)}$= co-variance of traits x and y, σ_x^2 = variance of x and σ_y^2 = variance of y.

RESULTS AND DISCUSSION

The crop season and test locations

The two locations received more or less similar amount of rainfall with different pattern of distribution but Ambo was more humid than Ginchi (**Figs. 1A, 1B**). It was witnessed that more or less the weather variables recorded did not deviate much from the long-term trends at both locations (data not shown), indicating that the present findings will be reproducible in other seasons. The physicochemical properties of the soils from the two test locations, Ambo and Ginchi, showed equal level of low nitrogen contents (0.103%) but high levels of K, Ca, Mg, Na and Fe (Jones 2003) with variable amounts. The levels of exchangeable cations were also high with pH values more or less closer to neutral. The level of soil phosphorus was high at Ambo and low at Ginchi (**Table 1**). Similar results were reported from previous analysis of soils from the same locations (Dibabe *et al*. 2001).

Relationships between characters

1. Symbiotic characters

There were significant positive association between a number of component characters and symbiotic nitrogen fixation in shoot, grain and total biomass (**Table 3**). Nitrogen contents in the shoot, grain and total biomass consistently showed significant positive association with fixation in the respective component parts and with each other in possible pairs. Almost perfect positive correlation coefficients were observed particularly between shoot nitrogen content and shoot nitrogen fixation (r = 0.97), grain nitrogen content and grain nitrogen fixation (r = 0.95), grain nitrogen content and above ground biomass nitrogen fixation (r = 0.93) and biomass nitrogen content and biomass nitrogen fixation (r = 0.90). Nitrogen fixation in grain also revealed perfect positive association with biomass nitrogen fixation (r = 0.99). This indicated that the higher the tissue nitrogen content the higher will be the amount of nitrogen from fixation and *vice versa*. Existence of a perfect correlation between two traits may indicate that the two traits are conditioned nearly by the same set of genes and physiological mechanisms (Falconer 1989). Nitrogen fixation in shoot, grain and above

Table 3 Correlation coefficients (r) between symbiotic characters in 155 chickpea genotypes grown at two locations in Ethiopia.

Characters[1]	NI	NN	NDW	SNC	SNF	GNC	GNF	BMNC	BMNF	FNAE	GNY	SNY	BNY
NI	1.00												
NN	0.56**	1.00											
NDW	0.88**	0.60**	1.00										
SNC	0.31**	0.32**	0.21**	1.00									
SNF	0.33**	0.33**	0.24**	0.97**	1.00								
GNC	0.30**	0.33**	0.27**	0.57**	0.63**	1.00							
GNF	0.32**	0.34**	0.30**	0.51**	0.61**	0.95**	1.00						
BMNC	0.34**	0.37**	0.28**	0.82**	0.85**	0.93**	0.87**	1.00					
BMNF	0.34**	0.35**	0.31**	0.59**	0.70**	0.93**	0.99**	0.90**	1.00				
FNAE	0.16*	0.21**	0.20*	0.03	0.16*	0.57**	0.78**	0.41*	0.74**	1.00			
GNY	0.09	0.05	0.15	-0.08	0.02	0.32**	0.42**	0.19*	0.40**	0.53**	1.00		
SNY	0.21**	0.27**	0.23**	0.65**	0.64**	0.39**	0.39**	0.55**	0.45**	0.13	0.44**	1.00	
BNY	0.17*	0.18*	0.22**	0.30**	0.35**	0.42**	0.49**	0.42**	0.50**	0.41**	0.88**	0.81**	1.00
NHI	-0.07	-0.16*	-0.02	-0.68**	-0.59**	-0.07	0.04	-0.34**	-0.04	0.41**	0.47**	-0.55**	0.011

[1]NI = nodulation index, NN = no of nodules, NDW = nodule dry weight, SNC = shoot nitrogen content, SNF = shoot nitrogen fixation, GNC = grain nitrogen content, GNF = grain nitrogen fixation, BMNC = biomass nitrogen content, BMNF = biomass nitrogen fixation, FNAE = fixed nitrogen assimilation efficiency, GNY = grain nitrogen yield, SNY = shoot nitrogen yield, BNY = biomass nitrogen yield and NHI = nitrogen harvest index; ** = highly significant ($P \leq 0.01$), * = significant ($P \leq 0.05$)

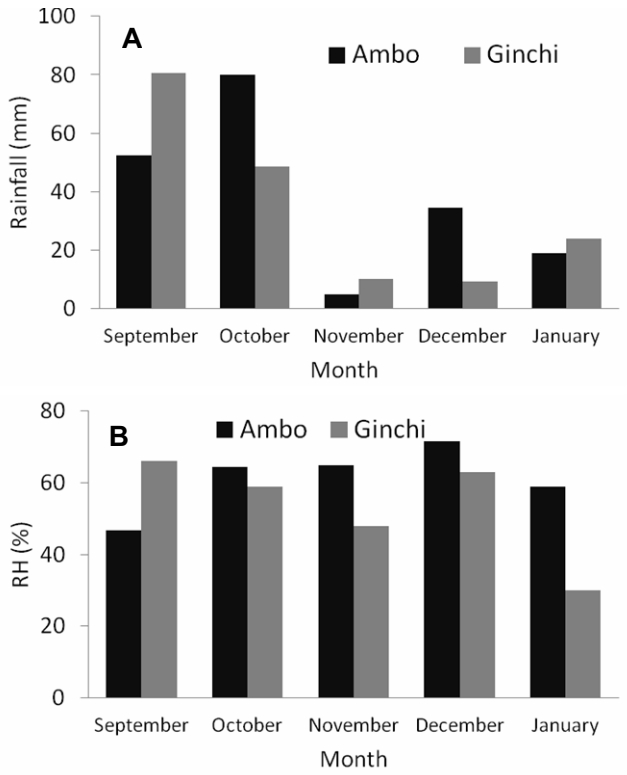

Fig. 1 (**A**) Rainfall (mm) and relative humidity (%) and (**B**) maximum and minimum temperatures (°C) at Ambo and Ginchi during the growing season.

ground biomass also showed significant positive associations with fixed nitrogen assimilation efficiency, indicating that better fixers of nitrogen were also better assimilators of the fixed nitrogen.

Better nodulation as expressed by number and weight of nodules and by the nodulation index showed consistently significant but relatively lower (as compared to tissue nitrogen content) positive associations with fixation in all component parts, i.e. shoot, grain and total above ground biomass (r = 0.24-0.35). Biabani *et al*. (2011) found that the amount of symbiotic nitrogen fixation in a global chickpea "mini-core" collection showed positively significant correlation coefficients with shoot (r = 0.21, P<0.01) and total biomass dry matter weight (r = 0.20, P<0.01) but not with nodule number (0.10) or weight (r = 0.13).

Despite the ease of observation (no need for laboratory analysis) and modest positive associations of nodule based traits (nodule number, weight and nodulation index) with the amount of symbiotic nitrogen fixation, the low level of genetic variation and heritability of those traits, as presented below, may not allow them to be surrogate trait for symbiotic nitrogen fixation. NHI was also positively associated with fixed nitrogen assimilation efficiency (r = 0.41) and grain nitrogen yield (r = 0.47). However, there were negatively significant associations between NHI and number of nodules (r = -0.16), NHI and shoot nitrogen content (r = -0.68), NHI and shoot nitrogen fixation (r = -0.59), NHI and above ground biomass nitrogen content (r = -0.34), and between NHI and shoot nitrogen yield (r = -0.55).

Characters with both perfect positive association and higher heritable variation include; grain nitrogen yield, NHI and fixed nitrogen assimilation efficiency. It is interesting to note that these characters also showed positive relationships with a number of other symbiotic and agronomic components (**Tables 3, 4**). In terms of total nitrogen output from a given genotype, it appeared that the gross amount of nitrogen as related to total biomass accumulation may be more important than *per se* concentration of nitrogen in plant tissue. Earlier reports also indicated the strong association between total nitrogen yield and dry matter production (r = 0.92) and between fixed nitrogen and dry matter production (r = 0.90) (Beck and Rupela 1996). However, these characters which possessed higher positive association and higher heritable variation are not easily observable as they need performing destructive sampling and laboratory analysis which, in turn, require additional labor, time and expense.

2. Agronomic characters

Grain yield was negatively associated with days to flowering (r = -0.45) and maturity (r = -0.24) but positively associated with grain filling period (r = 0.38). This may be attributed to the fact that long flowering and maturity durations might have directly reduced grain yield and indirectly have lowered other component characters like HI, grain production efficiency, and biomass production and economic growth rates. Longer days to flowering also negatively affected pod (r = -0.36) and seed (r = -0.39) setting. Similar results were reported from moisture stressed areas in Australia where higher yields were displayed by genotypes characterized by early flowering and rapidly setting pods (Berger *et al*. 2003). According to Wallace and Yan (1998), days to maturity usually correlated negatively with HI.

In other environments where moisture is not as such a limiting factor, different scenario may be observed in that late flowering and maturing genotypes may effectively exploit the available moisture and perform better than early genotypes that cannot fully exploit the moisture (Wallace and Yan 1998). For instance, in winter grown chickpea, where moisture use efficiency was better (Singh 1990), late maturing varieties gave better yields because of the pheno-

Table 4 Correlation coefficients (r) between agronomic characters in 155 chickpea genotypes grown at two locations in Ethiopia.

Characters[1]	SDWF	DTF	DTM	SFD	NP	NS	SDWM	BMWT	HI	PE	BPR	EGR	TSW
SDWF	1.00												
DTF	-0.23**	1.00											
DTM	-0.22**	0.54**	1.00										
GFD	0.15	-0.82**	0.03	1.00									
NP	0.01	-0.36**	-0.1	0.34**	1.00								
NS	-0.02	-0.39**	-0.06	0.42**	0.85**	1.00							
SDWM	0.22**	-0.05	-0.01	0.06	0.50**	0.43**	1.00						
BMWT	0.27**	-0.17*	-0.10	0.15	0.55**	0.49**	0.97**	1.00					
HI	0.11	-0.56**	-0.37**	0.41**	0.20*	0.27**	-0.20*	-0.01	1.00				
PE	0.28**	-0.71**	-0.24**	0.69**	0.55**	0.60**	0.51**	0.65**	0.60**	1.00			
BPR	0.29**	-0.23**	-0.22**	0.13	0.55**	0.48**	0.95**	0.99**	0.03	0.66**	1.00		
EGR	0.26**	-0.17*	-0.28**	0.02	0.47**	0.47**	0.71**	0.80**	0.40**	0.72**	0.82**	1.00	
TSW	0.27**	0.21**	-0.05	-0.27**	-0.58**	-0.67**	0.16*	0.15	-0.14	-0.07	0.15	0.16*	1.00
YLD	0.29**	-0.45**	-0.24**	0.38**	0.57**	0.59**	0.67**	0.79**	0.53**	0.92**	0.81**	0.93**	0.04

[1]SDWF = early vigor, DTF = days to 50% flowering, DTM = days to 90% maturity, GFD = grain filling duration, NP = no of pods, NS = no of seeds, SDWM = shoot dry weight at maturity, BMWT = biomass weight, HI = harvest index, PE = production efficiency, BPR = biomass production rate, EGR = economic growth rate, TSW = thousand seed weight and YLD = grain yield; ** = highly significant ($P \leq 0.01$), * = significant ($P \leq 0.05$)

Table 5 Correlation coefficients (r) between symbiotic and agronomic characters in 155 chickpea genotypes grown at two locations in Ethiopia.

Characters[1]	NI	NN	NDW	SNC	SNF	GNC	GNF	BMNC	BMNF	FNAE	SNY	GNY	BNY	NHI
SDMWF	-0.12	0.11	0.15	-0.14	-0.08	0.03	0.11	0.04	0.10	0.24**	0.06	0.28**	0.21**	0.20*
DTF	-0.16*	0.03	-0.15	0.17*	0.06	-0.14	0.30**	-0.03	0.28**	-0.53**	0.07	0.46**	0.27**	0.55**
DTM	-0.15	0.00	-0.17*	0.12	0.02	-0.14	-0.28**	-0.05	-0.27**	-0.45**	0.09	-0.26**	-0.13	-0.37**
SFD	0.10	-0.02	0.07	-0.10	-0.04	0.09	0.19*	0.02	0.18*	0.35**	0.01	0.39**	0.26**	0.40**
NP	0.15	0.01	0.12	0.01	0.04	0.13	0.19*	0.10	0.18*	0.24**	0.39**	0.59**	0.59**	0.18*
NS	0.16*	-0.04	0.10	0.02	0.05	0.15	0.21**	0.11	0.20*	0.27**	0.35**	0.61**	0.58**	0.24**
SDMW	0.03	0.08	0.15	0.01	0.03	0.06	0.12	0.04	0.13	0.19*	0.74**	0.66**	0.82**	-0.15
BMWT	0.04	0.06	0.17*	-0.13	-0.08	0.01	0.09	-0.05	0.08	0.24**	0.63**	0.75**	0.82**	0.05
HI	-0.07	-0.16*	-0.06	-0.34**	-0.29**	-0.07	0.03	-0.20*	0.01	0.29**	-0.37**	0.46**	0.11	0.82**
PE	0.02	-0.08	0.07	-0.29**	-0.20*	-0.02	0.14	-0.14	0.12	0.43**	0.19*	0.86**	0.66**	0.59**
BPR	0.06	0.05	0.18*	-0.15	-0.09	0.01	0.11	-0.05	0.10	0.28**	0.60**	0.77**	0.82**	0.10
EGR	-0.06	-0.06	0.03	-0.25**	-0.19*	-0.06	0.06	-0.15	0.05	0.30**	0.34**	0.86**	0.74**	0.40**
TSW	0.23**	-0.03	-0.08	-0.20*	-0.19*	-0.22**	-0.18*	-0.24**	-0.18*	-0.02	-0.04	-0.05	-0.05	-0.06
YLD	-0.03	-0.06	0.05	-0.28**	-0.20*	-0.03	0.11	-0.14	0.09	0.39**	0.31**	0.93**	0.77**	0.52**

[1]Abbreviations of characters as given in **Tables 3** and **4**; ** = highly significant ($P \leq 0.01$), * = significant ($P \leq 0.05$)

logical advantages (Özdemir and Kardavut 2003). The present study also showed that genotypes with longer relative grain filling period (days to maturity more or less kept constant) were characterized by better pod (r = 0.34) and seed (r = 0.42) setting, HI (r = 0.41), grain production efficiency (r = 0.69) and, hence, grain yield (r = 0.38). However, grain-filling period was negatively associated with seed size and that may be due to the fact that the large-seeded genotypes are introduced genotypes which have relatively longer vegetative period at the expense of reproductive period.

Other component characters that positively influenced grain yield include pod (r = 0.57) and seed (r = 0.59) setting, shoot biomass (r = 0.67) and total biomass (r = 0.79), HI (r = 0.53), grain production efficiency (r = 0.92), biomass production (r = 0.81), and economic growth (r = 0.93) rates. It is generally believed that higher biomass combined with higher HI is the major physiological and genetic components of grain yield (Wallace and Yan 1998). In this study, grain yield was not significantly influenced by seed size (r = 0.04), indicating that there is an independent genetic control between the two traits and that improvement in any one of the two would have little effect on the other. Yucel *et al*. (2006) also found a positive association between grain yield and number of pods and seeds but did not show any relationship between grain yield and seed size. Number of pods and seeds were also positively associated with a number of other characters but negatively related with seed size that may be because of the expected reciprocal compensation (Wallace and Yan 1998). The physiological parameters including shoot and biomass dry weight, HI, grain production efficiency, and biomass production and economic growth rates showed positive relationship among themselves in most of the cases (**Table 4**).

3. Symbiotic vs. agronomic characters

Grain yield showed negative relationship with increased shoot nitrogen content (r = -0.28) and shoot nitrogen fixation (r = -0.20). This implies that improving protein content of the shoot as a source of feed may have negatively influenced grain yield. However, grain and above-ground biomass nitrogen contents and nitrogen fixation did not show significant relationship with grain yield, indicating independent genetic control between the two traits and grain yield. On the other hand, correlation coefficients showed that grain yield could be significantly increased by increasing fixed nitrogen assimilation efficiency (r = 0.39), grain nitrogen yield (r = 0.93), shoot nitrogen yield (r = 0.31), biomass nitrogen yield (r = 0.77), and NHI (r = 0.52). On the contrary, increments in shoot and above ground biomass did not influence the amount of symbiotic nitrogen fixation in all component parts of the shoot, grain and total above-ground biomass.

Seed size had either negative or little relation with most of the symbiotic characters measured in this study with the exception of nodulation index which showed positive correlation (r = 0.23). This indicates that efforts to increase symbiotic nitrogen fixation may sometimes result in the reduction of seed size and *vice versa*. Biomass nitrogen fixation, fixed nitrogen assimilation efficiency, shoot, grain and biomass yields, and NHI showed significant positive associations with a number of agronomic yield components, and either non-siginificant or negative association with other components (**Table 5**). Generally, it can be concluded that symbiotic characters can make better selection criteria for grain yield than agronomic characters can do for symbiotic nitrogen fixation in all components.

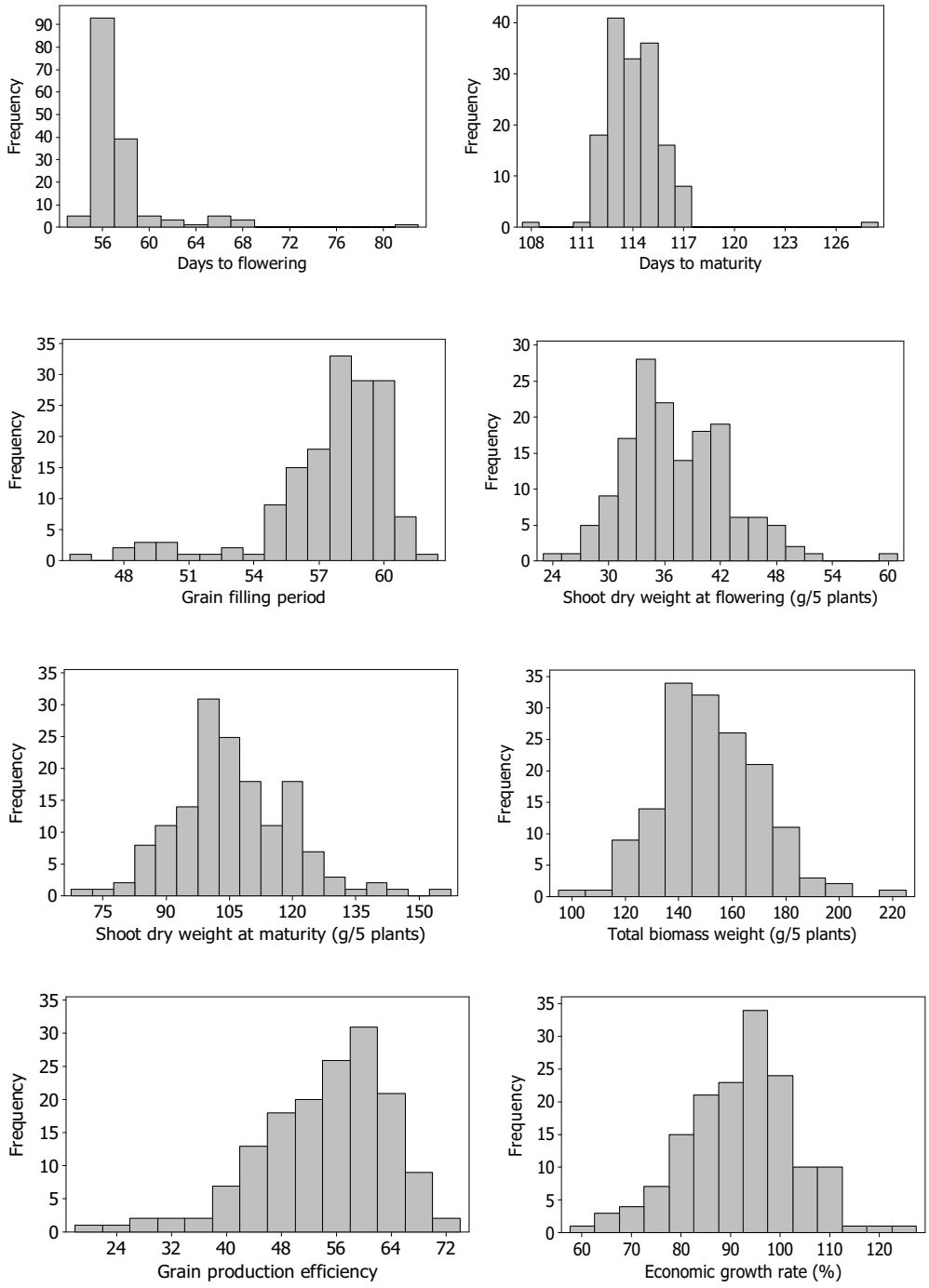

Fig. 2 Frequency distributions for 10 phenological and some physio-agronomic characters in 155 chickpea genotypes evaluated at two locations in Ethiopia.

Genetic variation

Differences among genotypes, locations and genotype by location interaction effects were significant for a number of characters. A number of accessions also better performed over the improved genotypes for both symbiotic and agronomic characters. The amount of fixed nitrogen ranged from 13-49% in foliage, 30-44% in grain and 28-40% in total above ground biomass. Grain yield performance varied from 31-70 g five plants^{-1} and seed size from 82-288 g/1000 seeds (data not shown).

The populations of chickpea genotypes studied here had normal distributions for most of the symbiotic and agronomic traits with only some levels of skewness for days to flowering and grain filling period, nitrogen fixed in grain and biomass, and fixed nitrogen assimilation efficiency (**Figs. 2-4**). This may be an indication that the Ethiopian chickpea germplasm accessions have normally distributed variation, and the individuals evaluated in this study represented the random samples of the Ethiopian chickpea germplasm collections (Welsh 1981). It is expected that, among genetically diverse natural population, the distribution of traits is usually normal (Simpson and Sedjo 1998).

Among the attributes of symbiotic nitrogen fixation, location and genotype by location interaction effects accounted for the largest part of the total variation for number of nodules, nodule dry weight, nodulation index and shoot and biomass nitrogen yields. Conversely, genotypic effects accounted for the largest part of the total variation for tissue nitrogen and protein contents and symbiotic nitrogen fixation in shoot, grain and biomass. The proportion of variation contributed by the genotypes was far less than that contributed by location and genotype by location interaction effects in shoot and above ground biomass dry weights and for biomass production and economic growth rates (**Fig. 5**). The greater proportion of location and genotype by location interaction variances relative to genotypic variance may suggest that selection in these traits would be less effective

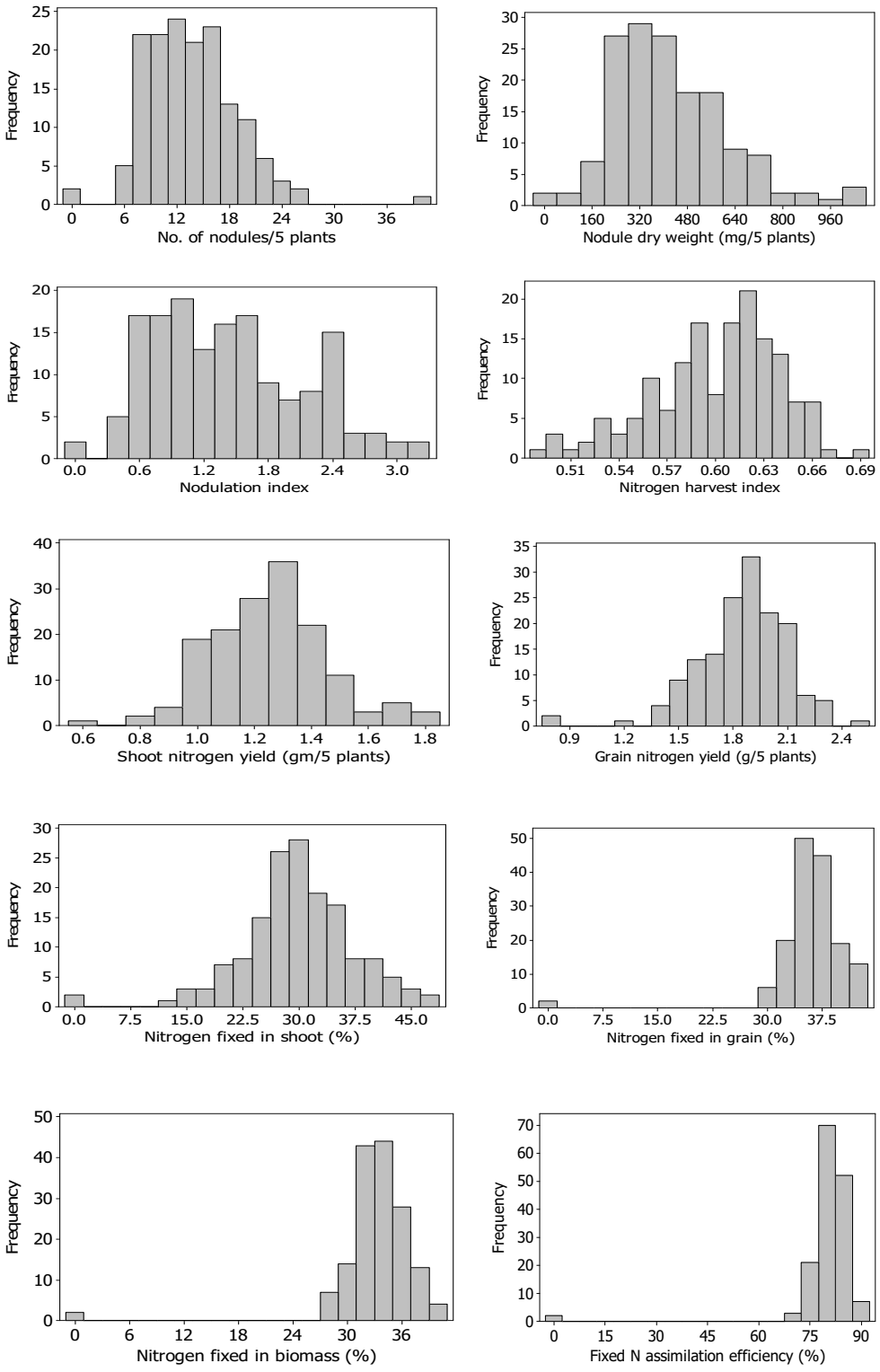

Fig. 3 Frequency distributions for 12 symbiotic characters in 155 chickpea genotypes evaluated at two locations in Ethiopia.

as compared to traits in which the largest proportion of the variation was contributed by the genotypes.

Broad-sense heritability and expected genetic gains from selection for traits of symbiotic and agronomic significance are presented in **Fig. 6**. From among the symbiotic characters, the highest broad-sense heritability values ranging from 80-91% were obtained for fixed nitrogen assimilation efficiency, and above ground biomass and seed nitrogen fixation. Shoot, seed and biomass nitrogen contents showed slightly better broad-sense heritability values ranging from 54-65%. As derivative traits, values of broad-sense heritability and expected genetic gains from selection for shoot, seed and biomass protein contents followed the same pattern (but 6.25 times in magnitude) as nitrogen contents of the same components. All characters related to nodulation (nodule number, nodule dry weight and nodulation index) generally exhibited relatively lower genotypic variation. Similarly, shoot and above ground biomass nitrogen yield had slightly better genotypic components of variation than nodulation but still very low as compared to the variation of location and genotype by location interaction components. The same traits obviously showed the least levels of broad-sense heritability values of 14% and 13%, respectively. This entails the need for evaluation of additional germplasm materials for these traits.

Traits with the highest expected genetic gains from selection at 5% selection intensity as percent of mean include fixation in shoot, seed and biomass and nitrogen

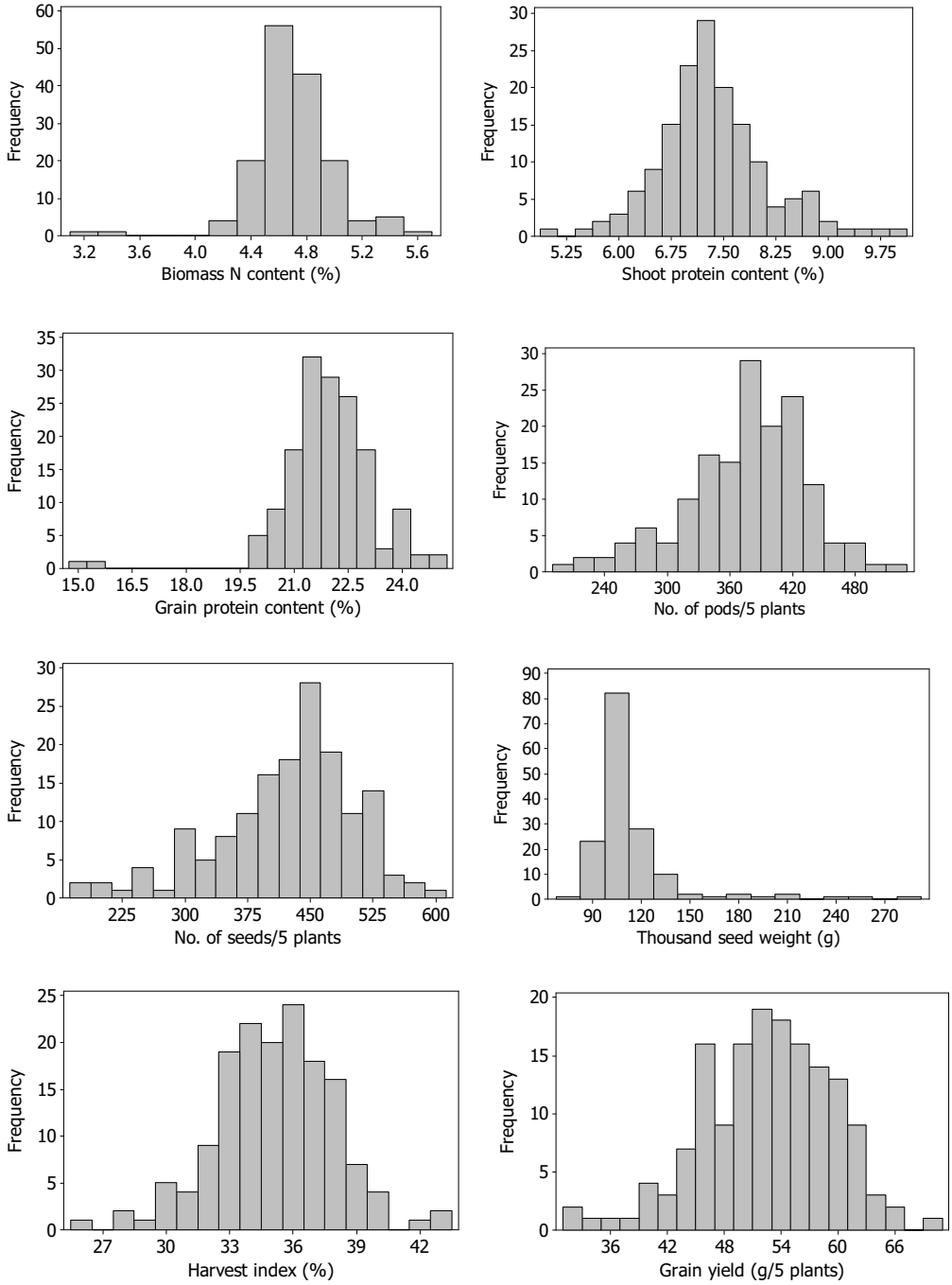

Fig. 4 Frequency distributions for 10 agronomic characters in 155 chickpea genotypes evaluated at two locations in Ethiopia.

assimilation efficiency with values ranging from 34-58%. Even though shoot and biomass nitrogen contents showed high heritable variations, values for expected genetic gains from selection were low (**Fig. 6**). Seed size and days to flowering showed the highest broad-sense heritability of 88% and 84%, respectively. Seed fill duration, number of seed and grain production efficiency showed modest broad-sense heritability values of 51 to 54%. Grain yield, HI, number of pods per plant and days to maturity had relatively lower heritability values ranging from 33-41%.

Other characters including shoot and biomass dry weights and biomass production and economic growth rates had the least heritability values of 11-18%. Seed size revealed the highest expected genetic gain from selection of 57% at 5% selection intensity. The highest broad-sense heritability and expected genetic gain from selection associated with seed size in this study may be attributed to the existence of uniquely large-seeded introduced materials from ICRISAT and ICARDA. Expected gains from selection in genotypes tested here revealed that number of pods and seeds, grain production efficiency and grain yield could be improved by about 20-38% (**Fig. 6**).

The Z-test showed significant differences between means of the selected subsets of the 5% best genotypes ($\overline{X}$) and the population parameters (μ) for all traits at phenotypic level. Comparison of the average performance for respective characters of the selected subsets of the 5% best genotypes with the average performances of the whole population for symbiotic characters revealed possibilities for different level of improvements through selection, ranging from 11% for fixed nitrogen assimilation efficiency to 119% for nodule dry weight. Above ground biomass nitrogen fixation can be improved by 18%. Likewise, possible improvements through selection for agronomic characters, disregarding phenological characters, also ranged from 17% for HI to 93% for seed size but the highest possible gain in the latter may be only due to existence of introduced varieties in the test genotypes. Grain yield can be improved by 23% (**Table 6**). This indicated that the selected accessions were not true representatives of the

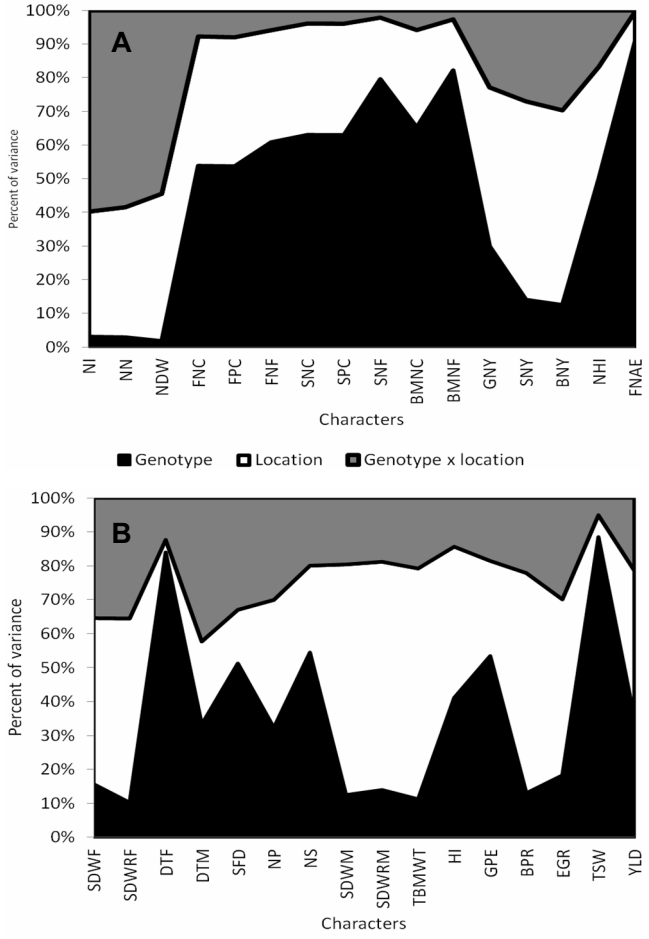

Fig. 5 Proportional contribution of components of variation (genotypic, location and genotype by location interaction effects) to the total variability in 155 chickpea genotypes for (**A**) symbiotic and (**B**) agronomic characteristics. SDWRF = shoot dry weight ratio at flowering, SDWRM = shoot dry matter weight ratio at maturity, and other abbreviations of characters as given in **Tables 3** and **4**.

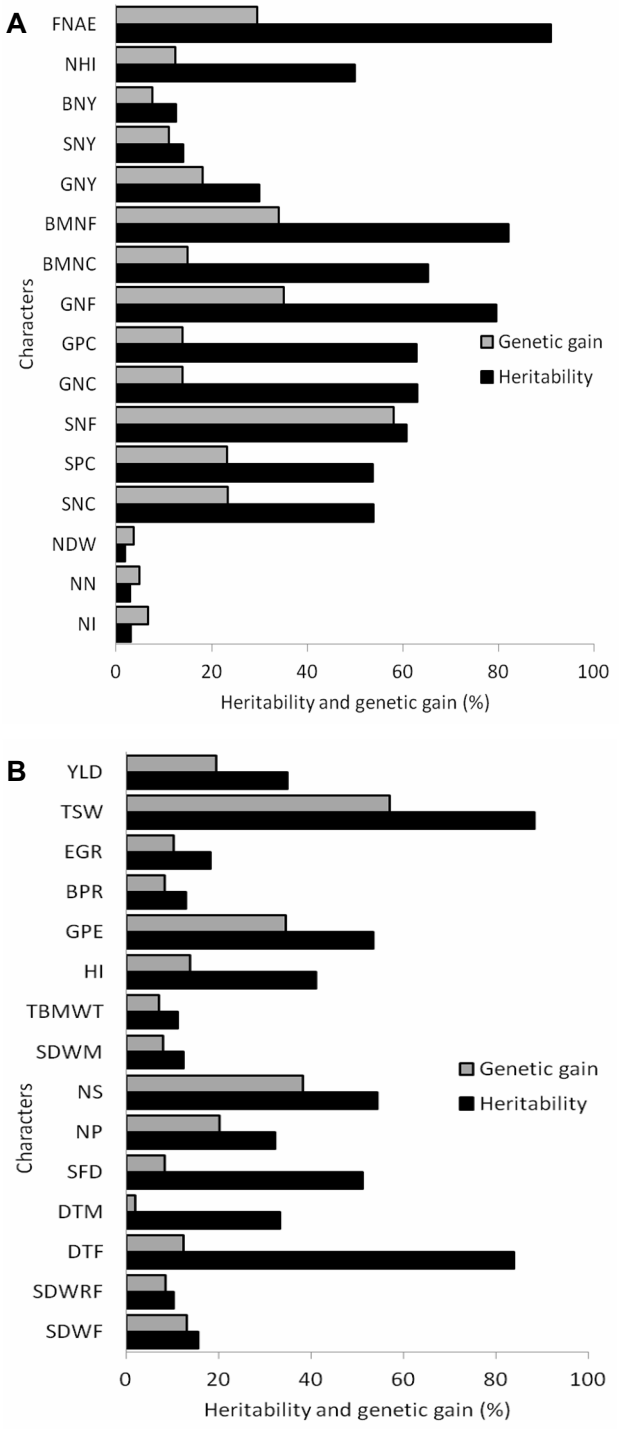

Fig. 6 Estimates of broad-sense heritability (h^2) and expected genetic advance (GA) from selection in 155 chickpea genotypes for (**A**) symbiotic and (**B**) agronomic characteristics. SPC = shoot protein content, GPC = grain protein content, SDWRF = shoot dry weight ratio at flowering, and other abbreviations of characters as given in **Tables 3** and **4**.

population and that almost all characters effectively responded to phenotypic selection (Singh 2001).

Genetic enhancement of these accessions to increase the frequency of best performing genes through further intra-accession selections would be expected to result in more promising lines. Biabani *et al*. (2011) also evaluated 39 accessions from the chickpea global core collection and found similar results. Nevertheless, it may be assumed that the effects of location and genotype by location interaction were found to influence traits with low broad-sense heritability and expected genetic gains from selection more strongly than the genetic effects. According to Singh (2005), selection for a trait with a high heritability (80% or more) should be fairly easy. There would be a close correspondence between genotype and phenotype due to a relatively smaller contribution of environment to the phenotype. But selection could be difficult or virtually impractical for a trait with low heritability, say less than 40% due to the environment concealing genotypic effects. Thus, it would be very difficult, to improve symbiotic traits like number of nodules, nodule dry weight and nodulation index through selection within the genotypes evaluated in this study. Likewise, it would be difficult to improve agronomic characters like shoot and biomass dry weight, biomass production and economic growth rates in these genotypes because of stronger environmental influence.

CONCLUDING REMARKS

This study showed that even though it may be difficult to get accessions that readily combine all desirable attributes into a single genotype, breeding for combined performances of symbiotic and agronomic attributes is possible through genetic manipulation. The level of genetic variation was within the level that permits effective selection in some of the symbiotic and agronomic traits measured.

Improving symbiotic nitrogen fixation may be more difficult than improving grain yield due to lack of positively associated traits with higher heritability that is easily observable during selection. For improving symbiotic nitrogen fixation, it appeared that increased nitrogen yield, NHI and fixed nitrogen assimilation efficiency with increased above ground biomass is more important than increasing the *per se* concentration of nitrogen in plant tissue. But as most of these traits are not easily observable, there is a need to

Table 6 Comparison of the mean performances of selected subsets ($\bar{x}$) of the 5% best accessions for symbiotic and agronomic characters with the average performances of the whole population (μ) of 155 chickpea genotypes.

Characters	Mean of selected genotypes ($\bar{x}$)	Population parameter (μ)	Change through selection ($\bar{x}$-μ)	Change as % of population parameter (μ)	Z
Symbiotic characters					
NI	2.93	1.43	1.50	104.90	6.05**
NN	25.50	13.21	12.29	93.04	6.66**
NDW	918.91	420.19	498.72	118.69	7.18**
SNC	1.48	1.18	0.30	25.42	6.59**
SNF	45.06	29.97	15.09	50.35	5.66**
GNC	3.90	3.51	0.39	11.11	5.37**
GNF	42.52	35.95	6.57	18.28	3.67**
BMNC	5.35	4.69	0.66	14.07	6.28**
BMNF	39.25	33.24	6.01	18.08	3.67**
FNAE	88.91	80.28	8.63	10.75	2.46*
GNY	2.31	1.85	0.46	24.86	5.12**
SNY	1.73	1.25	0.48	38.40	6.59**
BNY	3.79	3.10	0.69	22.26	5.16**
NHI	0.67	0.60	0.07	11.67	5.03**
Agronomic characters					
SDWF	50.66	37.4	13.26	35.45	6.60**
NP	483	376	107.00	28.46	5.27**
NS	557	421	136.00	32.30	4.62**
SDWM	137.34	105.87	31.47	29.73	6.61**
BMWT	196	152	44.00	28.95	6.43**
HI	40.99	35.15	5.84	16.61	5.91**
GPE	68.75	54.12	14.63	27.03	4.31**
BPR	174	133	41.00	30.83	6.67**
EGR	114	92	22.00	23.91	5.52**
TSW	220.03	113.8	106.23	93.35	10.35**
YLD	64.67	52.53	12.14	23.11	4.97**

** = highly significant ($P \leq 0.01$) and * = significant ($P \leq 0.05$)

develop either laboratory facilities or other handy tools that enable the analysis of plant tissue nitrogen content. For improving grain yield, in addition to components with high positive association and heritable variation, phenological characters may also need to be manipulated for better matching with the moisture regime. However, the need for more specific studies on the role of physiological attributes in chickpea breeding in general is obvious.

Negative associations were found between some important symbiotic and agronomic traits like grain yield with shoot nitrogen content and fixation; HI, grain production efficiency, biomass production and economic growth rates with shoot, grain or biomass nitrogen contents or fixation. That means improvement made in any one of the traits may cause reduction in the other and *vice versa*. Some possible strategies may help overcome this problem. In-depth studies may be needed to investigate whether the genetic nature of this negative relationship is associated with gene linkage or pleiotrophic gene effects. The result would enable to break linkages by producing large number of segregants (Singh 2005) if linkage is the main cause of negative association. It may also be possible to accept some level of compromise between two negatively related characters in such a way that one character should be kept constant in order to improve the other. It is wise to consider application of separate breeding programs particularly when the two traits that are associated negatively are controlled by pleiotrophic gene effects. A few morpho-agronomic characters in chickpea are under gene linkage and pleiotropic effects as reviewed by Muehlbauer and Singh (1987).

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Appendix 1 Passport description of the test genotypes.

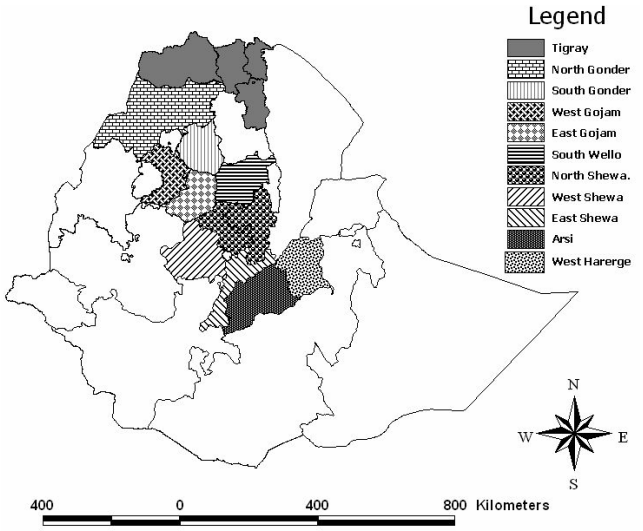
S. No.	Accession/genotype	Region	Zone	District	Altitude (masl)
1	Acc. No. 231327	Oromiya	Arsi	Merti	1540
2	Acc. No. 231328	Oromiya	Arsi	Jeju	1600
3	Acc. No. 209093	Oromiya	Arsi	Dodota Sire	1710
4	Acc. No. 208829	Oromiya	Arsi	Dodota Sire	1740
5	Acc. No. 209094	Oromiya	Arsi	Dodota Sire	1750
6	Acc. No. 209092	Oromiya	Arsi	Dodota Sire	1770
7	Acc. No. 209096	Oromiya	Arsi	Dodota Sire	1850
8	Acc. No. 209097	Oromiya	Arsi	Dodota Sire	1860
9	Acc. No. 209098	Oromiya	Arsi	Dodota Sire	1860
10	Acc. No. 41002	Oromiya	Arsi	Tena	2080
11	Acc. No. 207761	Oromiya	Arsi	Tena	2080
12	Acc. No. 207763	Oromiya	Arsi	Tena	2080
13	Acc. No. 207764	Oromiya	Arsi	Tena	2080
14	Acc. No. 41268	Amahara	E. Gojam	H. Ej Enese	1770
15	Acc. No. 41026	Amahara	E. Gojam	Hulet Ej Enese	2280
16	Acc. No. 41074	Amahara	E. Gojam	Hulet Ej Enese	2450
17	Acc. No. 41075	Amahara	E. Gojam	Hulet Ej Enese	2410
18	Acc. No. 41073	Amahara	E. Gojam	Hulet Ejenese	2400
19	Acc. No. 41076	Amahara	E. Gojam	Hulet Ej Enese	2470
20	Acc. No. 41021	Amahara	E. Gojam	Enarj Enawga	2510
21	Acc. No. 41027	Amahara	E. Gojam	Shebel Berenta	2450
22	Acc. No. 41222	Amahara	E. Gojam	Dejen	2460
23	Acc. No. 207734	Amahara	E. Gojam	Goncha Siso Enese	2560
24	Acc. No. 41103	Amahara	E. Gojam	Enemay	2570
25	Acc. No. 41320	Amahara	E. Gojam	Debay Telatgen	2400
26	Acc. No. 41029	Amahara	E. Gojam	Enarj Enawga	2880
27	Acc. No. 41015	Amahara	W. Gojam	Jabi Tehnan	2020
28	Acc. No. 41271	Amahara	W. Gojam	Adet	1880
29	Acc. No. 41272	Amahara	W. Gojam	Adet	1960
30	Acc. No. 41276	Amahara	W. Gojam	Adet	2230
31	Acc. No. 207745	Amahara	W. Gojam	Adet	2230
32	Acc. No. 41275	Amahara	W. Gojam	Adet	2240
33	Acc. No. 41277	Amahara	W. Gojam	Adet	2240
34	Acc. No. 207743	Amahara	W. Gojam	Adet	2240
35	Acc. No. 207744	Amahara	W. Gojam	Adet	2240
36	Acc. No. 41273	Amahara	W. Gojam	Adet	2300
37	Acc. No. 41274	Amahara	W. Gojam	Adet	2360
38	Acc. No. 207741	Amahara	W. Gojam	Adet	2360
39	Acc. No. 207742	Amahara	W. Gojam	Adet	2360
40	Acc. No. 41316	Amahara	N. Gonder	Gonder Zuria	1900
41	Acc. No. 41298	Amahara	N. Gonder	Gonder Zuria	1920
42	Acc. No. 41311	Amahara	N. Gonder	Dembia	1925
43	Acc. No. 41313	Amahara	N. Gonder	Gonder Zuria	1925
44	Acc. No. 41280	Amahara	N. Gonder	Gonder Zuria	1940
45	Acc. No. 41312	Amahara	N. Gonder	Gonder Zuria	1950
46	Acc. No. 41315	Amahara	N. Gonder	Gonder Zuria	1900
47	Acc. No. 41308	Amahara	N. Gonder	Dembia	2010
48	Acc. No. 41299	Amahara	N. Gonder	Gonder Zuria	1920
49	Acc. No. 41046	Amahara	N. Gonder	Chilga	2160
50	Acc. No. 41047	Amahara	N. Gonder	Chilga	2160
51	Acc. No. 41304	Amahara	N. Gonder	Dabat	2610
52	Acc. No. 41303	Amahara	N. Gonder	Wegera	2710
53	Acc. No. 41295	Amahara	S. Gonder	Fogera	1820
54	Acc. No. 41296	Amahara	S. Gonder	Kemekem	1850
55	Acc. No. 41289	Amahara	S. Gonder	Kemekem	1855
56	Acc. No. 41290	Amahara	S. Gonder	Kemekem	1880
57	Acc. No. 41284	Amahara	S. Gonder	Dera	1900
58	Acc. No. 41291	Amahara	S. Gonder	Kemekem	1900
59	Acc. No. 41297	Amahara	S. Gonder	Kemekem	1950
60	Acc. No. 41293	Amahara	S. Gonder	Kemekem	2040
61	Acc. No. 41019	Amahara	S. Gonder	Este	2500
62	Acc. No. 41048	Amahara	S. Gonder	Farta	2640
63	Acc. No. 41049	Amahara	S. Gonder	Farta	2640
64	Acc. No. 41053	Amahara	S. Gonder	Lay Gayint	3120
65	Acc. No. 41054	Oromiya	W. Harargie	Chiro	1500
66	Acc. No. 41052	Oromiya	W. Harargie	Mieso	1510
67	Acc. No. 209082	Oromiya	W. Harargie	Kuni	1680
68	Acc. No. 209083	Oromiya	W. Harargie	Kuni	1700
69	Acc. No. 209084	Oromiya	W. Harargie	Kuni	1700
70	Acc. No. 209091	Oromiya	W. Harargie	Habro	1730
71	Acc. No. 209087	Oromiya	W. Harargie	Kuni	1740
72	Acc. No. 209088	Oromiya	W. Harargie	Habro	1740
73	Acc. No. 209089	Oromiya	W. Harargie	Habro	1740

Appendix 1 (Cont.)

S. No.	Accession/genotype	Region	Zone	District	Altitude (masl)
74	Acc. No. 209090	Oromiya	W. Harargie	Habro	1740
75	Acc. No. 209081	Oromiya	W. Harargie	Girawa	2130
76	Acc. No. 41159	Oromiya	E. Shewa	Ada'a Chukala	1910
77	Acc. No. 41160	Oromiya	E. Shewa	Ada'a Chukala	1910
78	Acc. No. 41161	Oromiya	E. Shewa	Ada'a Chukala	1940
79	Acc. No. 207661	Oromiya	E. Shewa	Ada'a Chukala	1850
80	Acc. No. 207667	Oromiya	E. Shewa	Akaki	2180
81	Acc. No. 207666	Oromiya	E. Shewa	Akaki	2060
82	Acc. No. 41141	Oromiya	E. Shewa	Lome	2040
83	Acc. No. 207665	Oromiya	E. Shewa	Akaki	2060
84	Acc. No. 41134	Oromiya	E. Shewa	Akaki	2080
85	Acc. No. 41128	Oromiya	E. Shewa	Akaki	2130
86	Acc. No. 41168	Oromiya	E. Shewa	Ada'a Chukala	2150
87	Acc. No. 41129	Oromiya	E. Shewa	Akaki	2170
88	Acc. No. 41130	Oromiya	E. Shewa	Akaki	2190
89	Acc. No. 41110	Amara	N. Shewa	Kewet	1220
90	Acc. No. 207657	Amara	N. Shewa	Efratana Gidim	1400
91	Acc. No. 41111	Amahara	N. Shewa	Efratana Gidim	1400
92	Acc. No. 41106	Amahara	N. Shewa	Mafudmezezo Mojana	1820
93	Acc. No. 207658	Amahara	N. Shewa	Efratana Gidim	1400
94	Acc. No. 41142	Amahara	N. Shewa	Minjarna Shenkora	2290
95	Acc. No. 41207	Amahara	N. Shewa	Siyadebrina Wayuense	2580
96	Acc. No. 41215	Amahara	N. Shewa	Moretena Jiru	2640
97	Acc. No. 41216	Amahara	N. Shewa	Moretena Jiru	2640
98	Acc. No. 41066	Oromiya	N. Shewa	Wara Jarso	2550
99	Acc. No. 41011	Oromiya	N. Shewa	Gerar Jarso	2558
100	Acc. No. 41007	Oromiya	N. Shewa	Yaya Gulale	2670
101	Acc. No. 41008	Oromiya	N. Shewa	Yaya Gulale	2700
102	Acc. No. 41186	Oromiya	W. Shewa	Waliso Goro	1960
103	Acc. No. 209035	Oromiya	W. Shewa	Alem Gena	2010
104	Acc. No. 41176	Oromiya	W. Shewa	Ambo	2020
105	Acc. No. 41175	Oromiya	W. Shewa	Ambo	1970
106	Acc. No. 41174	Oromiya	W. Shewa	Ambo	2120
107	Acc. No. 209027	Oromiya	W. Shewa	Kersa Kondaltiti	2060
108	Acc. No. 41170	Oromiya	W. Shewa	Dendi	2160
109	Acc. No. 41171	Oromiya	W. Shewa	Dendi	2230
110	Acc. No. 41185	Oromiya	W. Shewa	Woliso Goro	2000
111	Acc. No. 209036	Oromiya	W. Shewa	Alem Gena	2220
112	Acc. No. 41190	Oromiya	W. Shewa	Woliso Goro	2080
113	Acc. No. 41195	Oromiya	W. Shewa	Becho	2160
114	Acc. No. 41197	Oromiya	W. Shewa	Becho	2120
115	Acc. No. 207150	Tigray	S. Tigray	Enderta	---
116	Acc. No. 207151	Tigray	S. Tigray	Enderta	---
117	Acc. No. 207563	Tigray	S. Tigray	Hintalo Wajirat	1960
118	Acc. No. 207564	Tigray	C. Tigray	Laelay Maychew	2150
119	Acc. No. 207894	Tigray	S. Tigray	Endamehoni	2600
120	Acc. No. 207895	Tigray	S. Tigray	Alaje	---
121	Acc. No. 213224	Tigray	E. Tigray	Wukro	2100
122	Acc. No. 219797	Tigray	C. Tigray	Laelay Maychew	2150
123	Acc. No. 219799	Tigray	C. Tigray	Laelay Maychew	1970
124	Acc. No. 219800	Tigray	C. Tigray	Adwa	2400
125	Acc. No. 219803	Tigray	W. Tigray	Tahtay Koraro	1880
126	Acc. No. 221696	Tigray	S. Tigray	Enderta	---
127	Acc. No. 41114	Amahara	S. Wello	Werebabu	1560
128	Acc. No. 212589	Amahara	S. Wello	Kalu	1600
129	Acc. No. 41113	Amahara	S. Wello	Kalu	1650
130	Acc. No. 207659	Amahara	S. Wello	Dessie Zuria	1950
131	Acc. No. 207660	Amahara	S. Wello	Dessie Zuria	1950
132	Acc. No. 41115	Amahara	S. Wello	Kutaber	2290
133	Acc. No. 225878	Amahara	S. Wello	Debresina	2420
134	Acc. No. 225873	Amahara	S. Wello	Debresina	2445
135	Acc. No. 225874	Amahara	S. Wello	Debresina	2450
136	Acc. No. 225877	Amahara	S. Wello	Kelala	2450
137	Acc. No. 207645	Amahara	S. Wello	Debresina	2510
138	Acc. No. 207646	Amahara	S. Wello	Debresina	2510
139	Acc. No. 225876	Amahara	S. Wello	Kelala	2540
140	ICC 5003*	India	---	---	---
141	ICC 4918	India	---	---	---
142	ICC 4948	India	---	---	---
143	ICC 4973	India	---	---	---
144	ICC 15996	ICRISAT	---	---	---
145	Shasho (ICCV 93512)	ICRISAT	---	---	---

Appendix 1 (Cont.)

S. No.	Accession/genotype	Region	Zone	District	Altitude (masl)
146	Arerti (FLIP 89-84C)	ICARDA	---	---	---
147	Worku (DZ-10-16-2)	ICRISAT	---	---	---
148	Akaki (DZ-10-9-2)	ICRISAT	---	---	---
149	Ejere (FLIP-97-263C)	ICARDA	---	---	---
150	Teji (FLI 97-266C)	ICARDA	---	---	---
151	Habru (FLIP 88-42c)	ICARDA	---	---	---
152	Natoli (ICCX-910112-6)	ICRISAT	---	---	---
153	ICC 19180	ICRISAT	---	---	---
154	ICC 19181	ICRISAT	---	---	---
155	PM 233 (155)	ICARDA	---	---	---



Appendix 2 Map of Ethiopia showing the approximate areas of origins (shaded region) of the 139 germplasm accessions. NB: all boundaries are approximate and nothing to do with political borders.