

Identification of Large-Seeded High-Yielding Stable Kabuli Chickpea Germplasm Lines for Use in Crop Improvement

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ABSTRACT

Seed size is an important trait in kabuli chickpea (*Cicer arietinum* L.). The kabuli chickpeas with a 100-seed weight of >40 g garner higher market price as they are preferred by consumers. The objective of this study was to evaluate a sample of chickpea landraces, breeding lines, and cultivars for morphological traits and yield and identify lines that are high yielding and exhibit stable performance across environments. We evaluated 65 large-seeded kabuli lines identified from the ICRISAT germplasm collection in three sets, 18 trials, and 13 environments for 22 qualitative and quantitative traits. Several highly significant correlations were observed in all three sets and a few large-seeded high-yielding lines with stable yield were selected. Two extra-large-seeded (100-seed weight > 50 g) lines ICC 17109 (Blanco Sinaloa 92), a breeding line, and ICC 17452 (CuGa 288), a landrace, both originating from Mexico showed high yield potential and were moderately stable across environments. The small-seeded control cultivars, though high yielding, were highly unstable, performing better only in favorable environments. The study has also shown that some vegetative characteristics were more sensitive to stress than yield and yield components, hence selection in unfavorable environments should be based on grain yield and its components.

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Abbreviations: b_1 , linear component; E, environment, h^2_b , broad-sense heritability; PC, principal component; PCA, principal component analysis; REML, residual (or restricted) maximum likelihood; $S^2_{d_1}$, nonlinear component; σ^2_e , residual (error) variance; σ^2_g , genotypic variance; σ^2_{ge} , genotype \times environment interaction variance.

GRAIN LEGUMES are a rich dietary source of good quality proteins, carbohydrates, fiber, and minerals (calcium, potassium, phosphorus, iron, zinc, and magnesium) that benefit the overall health of people, especially vegetarians in South Asia who consume pulses as a main source of dietary protein along with cereals. Chickpea (*Cicer arietinum* L.), which ranks second among edible pulses in global markets (Yadav et al., 2007), is widely cultivated in the Mediterranean, North Africa, the Middle East, and the Indian subcontinent. It is the most important legume in Asia, with about 90% of global area and 88% of production. The world cropping area under chickpea is about 11.56 million ha, with a total production of 8.78 million Mt and an average productivity of 0.76 Mt ha⁻¹ (FAO, 2008). The major chickpea producing countries include India, Pakistan, Iran, Turkey, Australia, Myanmar, Ethiopia, Canada, Mexico, Syria, the United States, Spain, Eritrea, and Bangladesh, of which India is the largest producer (7.54 million ha area and 5.75 million Mt production).

Chickpea has the inherent capacity to tolerate drought as it grows well under residual soil moisture after the cessation of rains. In addition, hair-like structures (trichomes) on its stem,

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leaves, and pods secrete acids that provide the first line of defense against pests, reducing the need for chemical sprays (Yadav et al., 2007).

The two major forms of chickpeas are desi and kabuli types, the classification of which is based on seed size, shape, and color. The small, angular shaped and colored seeds are known as desi types, while large, owl's head shaped and beige colored seeds are kabuli types. Desi seeds are usually dehulled and split to make dhal or flour (Besan) while Kabuli seeds are often cooked as whole grain. Seed size and color are important traits for trade. Consumers prefer the large-seeded types for whole seed consumption, confectionary products, salads, and savory meals (Regan et al., 2006). The extra large (>50 g 100-seed^{-1}) kabuli cultivars are sold at three times the price of desi and twice the price of kabuli types with a 100-seed weight from 25 to 40 g in India (Gaur et al., 2006).

The trade recognizes three groups based on seed diameter in Europe and Australia: large seeded (>9 mm), medium seeded (8–9 mm), and small seeded (7–8 mm). Kabuli chickpea seeds of more than 7 mm receive a premium of US \$50 per ton for each additional mm of diameter (Biçer, 2009). The seed lot must contain a large proportion of such seeds to get the extra price (Barker, 2007). The producers in Canada receive a weighted price based on seed size and uniformity (Yadav et al., 2007). Kabuli chickpea now accounts for 20% of world chickpea production (Regan et al., 2006).

Developing large-seeded high-yielding kabuli cultivars is an important breeding objective in most chickpea improvement programs, and identification of large-seeded kabuli germplasm lines is a prerequisite to initiate such programs. A large number of chickpea germplasm accessions (more than 98,000) are conserved in several genebanks. However, only a small proportion has been used in the national and international breeding programs. In India, which has a strong chickpea breeding program, 41% of the 126 cultivars released in the past four decades have Pb 7 (desi type) in their pedigree followed by IP 58, F 8, S 26 (all desi), and Rabat (kabuli; 34 g 100-seeds^{-1}) (Kumar et al., 2004). In the breeding program at ICRISAT, less than 1% of germplasm has been used in developing more than 3700 breeding lines during 1978–2008 (Upadhyaya et al., 2006b, 2009). Of the 92 germplasm lines used, only 19 were kabuli types, six of which had large seed size (>40 g 100-seed^{-1}). L 550, a small-seeded (20 g 100-seed^{-1}) kabuli cultivar was most frequently used (983 times) in the pedigree. One of the main reasons for low use of germplasm in breeding programs is the lack of information on traits of economic importance, which often show genotype \times environment interactions and require multi-environment evaluation. Several studies have been conducted on characterization and estimating of diversity, heritability, and genetic gain using a limited number of germplasm lines

(<50) in a single environment. In most such studies, except Singh et al. (1980), Gaur et al. (2006), and Upadhyaya et al. (2006a), who studied kabuli types, the germplasm was not classified as desi or kabuli types.

ICRISAT has a global collection of 20,140 chickpea accessions from 60 countries. The core (10% of the entire collection) (1956 accessions) (Upadhyaya et al., 2001) and mini core (10% of core or 1% of the entire collection) (211 accessions) (Upadhyaya and Ortiz, 2001) collections, which represent the diversity of the entire collection, have been reported to enhance the utilization of chickpea germplasm in crop improvement. The core and mini core approaches have been very useful in identifying new sources of variation (reviewed in Upadhyaya et al., 2009). In chickpea, the large-seeded kabuli types produce a lower yield than the small-seeded kabuli cultivars. Liu et al. (2003) reported that the large-seeded cultivars produced $<90\%$ yield per unit area of the small-seeded cultivars, mainly due to a lower number of pods per unit area, greater number of sterile (empty) pods, and a fewer number of seeds per pod than the small-seeded cultivars. Yadav et al. (2004) observed a negative correlation between yield and seed size under water-limiting environment. Large-seeded, high-yielding stable germplasm lines are not available for use in breeding programs. We selected 16 large-seeded kabuli chickpea lines from the core collection in 1999–2000 (Set-I) and 33 from the reserve collection in 2003–2004 (Set-II). Additionally, we selected 16 lines (Set-III) using 2004–2005 evaluation data on 335 newly assembled kabuli germplasm from the United States. Due to differences in seasons in identification of these lines, their evaluation for agronomic traits was continued separately in three sets at five to seven environments. The aim of this study was to identify large-seeded, high-yielding, stable kabuli germplasm lines for use in chickpea improvement.

MATERIALS AND METHODS

The present study consisted of 65 large-seeded kabuli chickpea germplasm lines, which included 45 landraces, 17 breeding lines, three advance cultivars, and four control cultivars (Table 1). These lines were selected based on seed size and originated from 11 major chickpea growing countries from different regions of the world. All the test entries in the three sets had smooth seed surfaces except ICC 14926 and ICC 16674 in Set-I, which had rough surfaces. Most of the entries were semi-erect types, with few exceptions: ICC 17456, an erect type, in Set-III, ICC 7344 in Set-I, ICC# 7345, 11295, 11883, 14193, 14361, and 16750 in Set-II, and ICC# 19189 and 19191 in Set-III were of semispreading types. Surutato 77 (ICC 19195), Mocarito-88 (ICC 19196), and W6 17604 (ICC 17459) are advanced cultivars from Mexico; thirteen (ICC# 7344, 14199, and 17109 in Set-I and ICC# 7345, 7346, 7347, 12033, 14195, 14196, 14197, 14201, 14214, and 14361 in Set-II) from Mexico and four from Chile were breeding lines (ICC 16674 in Set-I and ICC# 11815, 11816, 11821 in Set-II), while the remaining were landraces from different countries. The four common control cultivars

Table 1. Country of origin and biological status of large-seeded kabuli chickpea germplasm accessions included in this study.

Germplasm line	Identity	Country of origin	Biological status	Germplasm line	Identity	Country of origin	Biological status
Set-I				Set-II			
ICC 7344	DKN-1	Mexico	Breeding line	ICC 6187	NEC 54	Spain	Landrace
ICC 8155	NEC 2302	United States	Landrace	ICC 6210	NEC 78	Spain	Landrace
ICC 11303	708017	Chile	Landrace	ICC 6243	NEC 116	Tunisia	Landrace
ICC 12498	Sample 2	Turkey	Landrace	ICC 7345	Bremizado	Mexico	Breeding line
ICC 13787	BG 1-392	Spain	Landrace	ICC 7346	Culiacancito (860)	Mexico	Breeding line
ICC 14190	P 57-1	India	Landrace	ICC 7347	Guamuchil (916)	Mexico	Breeding line
ICC 14194	STO Domingo 82	Mexico	Landrace	ICC 7713	NEC 56	Spain	Landrace
ICC 14199	Varietad breve blanco	Mexico	Breeding line	ICC 8151	NEC 2298	United States	Landrace
ICC 14204	120-TM	Mexico	Landrace	ICC 8156	NEC 2303	United States	Landrace
ICC 14205	125-TBB	Mexico	Landrace	ICC 11295	No. 4	United States	Landrace
ICC 14926	PI 110408	Italy	Landrace	ICC 11745	INIA 24	Chile	Landrace
ICC 15331	KC 215085-1	Iran	Landrace	ICC 11815	INIA 103	Chile	Breeding line
ICC 16670	Tammany	United States	Landrace	ICC 11816	INIA 104	Chile	Breeding line
ICC 16674	INIA 110-1	Chile	Breeding line	ICC 11821	INIA 110	Chile	Breeding line
ICC 16744	Garo de bico	Portugal	Landrace	ICC 11883	Spanish cultivar	Spain	Landrace
ICC 17109	Blanco Sinaloa 92	Mexico	Breeding line	ICC 12033	F4 Line triple cross	Mexico	Breeding line
Set-III				ICC 13821	41121	Ethiopia	Landrace
ICC 17450	CuGa-290	Mexico	Landrace	ICC 14188	CG 276-1	Mexico	Landrace
ICC 17452	CuGa-288	Mexico	Landrace	ICC 14193	Rar TA RBB-1 II GAB-S11-M-M	Mexico	Landrace
ICC 17456	CuGa-208	Mexico	Landrace	ICC 14195	11-M-M-TARA-5II GAM-S1-M-6-M-M	Mexico	Breeding line
ICC 17457	CuGa-137	Mexico	Landrace	ICC 14196	6-M-M TARM-9 120 TM	Mexico	Breeding line
ICC 17458	CuGa-259	Mexico	Landrace	ICC 14197	TA RM-9 120 TM Garbanzo	Mexico	Breeding line
ICC 17459	W6 17604	Mexico	Advance cultivar	ICC 14201	Varietad union 13	Mexico	Breeding line
ICC 18591	W6 17607	Mexico	Landrace	ICC 14203	120-TBB	Mexico	Landrace
ICC 19188	M 89-13	Mexico	Landrace	ICC 14206	650 A Gordo lechoso	Mexico	Landrace
ICC 19189	Dorado	Mexico	Landrace	ICC 14207	650 A Pedro sevillano	Mexico	Landrace
ICC 19190	CA 188 178	Mexico	Landrace	ICC 14214	956-14II 26GA-76-1-M	Mexico	Breeding line
ICC 19191	CuGa-289	Mexico	Landrace	ICC 14361	1030-91 II 18GA-M-M-M-M-1	Mexico	Breeding line
ICC 19192	CuGa-291	Mexico	Landrace	ICC 16750	CHK 1377	Portugal	Landrace
ICC 19193	CuGa-257	Mexico	Landrace	ICC 16790	CHK 2018; 02504	Portugal	Landrace
ICC 19194	CuGa-141	Mexico	Landrace	ICC 16803	CHK 2035; 02552	Portugal	Landrace
ICC 19195	Surutato-77	Mexico	Advance cultivar	ICC 16819	CHK 2068	Portugal	Landrace
ICC 19196	Mocorito-88	Mexico	Advance cultivar	ICC 16821	CHK 2511	Portugal	Landrace
Common Controls:							
ICCV 2	Sweta	India	Released cultivar	JGK 1	–	India	Released cultivar
KAK 2	PKV-Kabuli-2	India	Released cultivar	L 550	–	India	Released cultivar

released in India in all the three sets were ICCV 2, JGK 1, KAK 2, and L 550. L 550 is a semi-erect, small-seeded kabuli type tolerant to root knot nematodes (Dua et al., 2001). ICCV 2 (Sweta) is an early maturing, semispreading, small-seeded kabuli type, resistant to Fusarium wilt (Kumar et al., 1985). KAK 2 (PKV-Kabuli-2) is a semi-erect type with large seed size that is resistant to Fusarium wilt and adapted to irrigated conditions (Zope et al., 2002). JGK-1 is a semi-erect kabuli type with large seed size, resistance to Fusarium wilt, and tolerance to pod borer (*Helicoverpa armigera* Huebn.) (Gaur et al., 2004).

Set-I was evaluated in seven environments (E), Set-II in five environments, and Set-III in six environments at Patancheru, India, (18° N, 78° E, 545 m above sea level), mostly under irrigated conditions, except E5 and E7 in Set-I, E11 in Set-II and E11 and E13 in Set-III, which were rainfed environments (Table 2). All the experiments were planted in vertisols using a spacing of 30 cm between rows and 10 cm between plants on broad beds, except for five trials in Set-I, which had a spacing

of 60 cm between rows and 10 cm between plants on ridges. All the experiments were conducted under receding moisture during the post-rainy season and the experiments were sown in the last week of October each year. Uniform depths of sowing and optimum plant population levels were maintained. The experiments received 16 kg N and 46 kg P₂O₅ ha⁻¹ as supplemental fertilizer and other crop husbandry practices were followed as required. As rainfall was deficient in all the seasons, a presowing irrigation and two protective irrigations (5 cm water per irrigation), one at initiation of flowering and the other during pod development, were given in the experiments under irrigation (I). For the experiments under rainfed (RF) condition only presowing irrigation was provided (Table 2).

Five competitive representative plants were selected randomly in each plot to record observations on plant height (cm), plant width (cm), and the number of basal primary branches, apical primary branches, basal secondary branches, apical secondary branches, tertiary branches, and pods per plant. The data

Table 2. Weather data for the test environments (1999–2000 to 2008–2009) at ICRISAT Patancheru, India.

Environment (E)	Season	Set I†	Set II	Set III	Weekly weather parameters during the cropping period											
					Rain fall (mm)			Evaporation (mm)			Max. temperature (°C)		Min. temperature (°C)		Bright sunshine hrs	
					min.	max.	total	min.	max.	total	min.	max.	min.	max.	min.	max.
E1	1999–2000	I†	–	–	0	14.8	20	3.0	8.7	603	25.6	35.0	5.8	20.7	0.6	10.7
E2	2000–2001	I	–	–	0	14.2	23	1.3	8.0	595	21.4	34.2	7.6	20.4	0.0	11.0
E3	2001–2002	I	–	–	0	22.4	31	2.1	7.8	546	24.8	33.2	6.2	20.5	1.5	10.8
E4	2003–02004	I	I	–	0	32.6	65	1.3	7.4	493	23.2	31.4	8.4	21.2	1.1	10.9
E5	2003–2004	RF†	–	–	0	32.6	65	1.3	7.4	493	23.2	31.4	8.4	21.2	1.1	10.9
E6	2004–2005	I	–	–	0	28.9	34	2.4	8.3	549	24.9	35.0	8.2	21.3	0.0	10.8
E7	2004–2005	RF	–	–	0	28.9	34	2.4	8.3	549	24.9	35.0	8.2	21.3	0.0	10.8
E8	2005–2006	–	I	I	0	79.2	118	1.2	6.9	507	22.8	32.8	6.7	22.7	0.0	10.8
E9	2006–2007	–	I	I	0	10.0	25	0.7	5.8	534	24.1	32.5	8.6	21.7	0.1	10.2
E10	2007–2008	–	I	I	0	51.4	101	2.6	6.9	559	26.5	32.4	7.3	22.3	0.0	10.5
E11	2007–2008	–	RF	RF	0	51.4	101	2.6	6.9	559	26.5	32.4	7.3	22.3	0.0	10.5
E12	2008–2009	–	–	I	0	12.4	27	0.8	7.4	531	23.6	34.4	9.5	22.0	0.0	10.6
E13	2008–2009	–	–	RF	0	12.4	27	0.8	7.4	531	23.6	34.4	9.5	22.0	0.0	10.6

†I, irrigated.

*RF, rainfed.

on days to 50% flowering, flowering duration (days), grain filling duration (days), days to maturity, 100-seed weight (g), grain yield (kg ha⁻¹), and productivity (kg ha⁻¹ d⁻¹) were recorded on plot basis following IBPGR, ICRISAT, and ICARDA (1993) descriptors. Data on each set was analyzed separately for each environment using residual (or restricted) maximum likelihood (REML; Patterson and Thompson, 1971) in GenStat 12 (available at <http://www.vsnl.co.uk>; verified 29 Sept. 2010). Pooled analysis for each set separately and pooled for all sets was performed using REML Meta analysis (DerSimonian and Laird, 1986; Hardy and Thompson, 1996; Whitehead, 2002). Significance of differences among seasons was tested using Wald (1943) statistics. Best linear unbiased predictors (BLUPs) (Schönfeld and Werner, 1986) were determined for all quantitative traits. The broad-sense heritability (h^2_b) was estimated for each environment separately and for pooled data for each set. Stability analysis based on Eberhart and Russell's (1966) model was performed to identify stable genotypes in each set separately. Principal component analysis (PCA) was performed for dimensional reduction and to identify the traits important in explaining variation. Cluster analysis was done following the minimum variance method of Ward (1963) to group together similar genotypes based on principal component (PC) scores. Means and variances of clusters were tested for significance following the Newman-Keuls procedure (Newman, 1939; Keuls 1952) and Levene (1960) test, respectively. Phenotypic correlations among all the traits were calculated for each environment and for pooled data.

RESULTS AND DISCUSSION

For the purpose of summarization of results and discussion, the traits studied were grouped into three broad categories based on the life cycle of the chickpea plant: (i) vegetative traits including plant height (cm), plant width (cm), and number of basal primary branches, apical primary branches, basal secondary branches, apical secondary

branches, and tertiary branches; (ii) reproductive traits including days to 50% flowering, flowering duration (days), days to maturity, and grain filling duration (days), and (iii) yield and yield component traits including the number of pods per plant, 100-seed weight (g), grain yield (kg ha⁻¹), and productivity (kg ha⁻¹ d⁻¹).

The range and mean of different traits revealed that the ranges were similar across traits in all sets while the mean values varied considerably (Table 3). Means of all the three sets differed significantly ($p = 0.05$) for plant width, days to maturity, number of basal primary and secondary branches, number of apical primary and secondary and tertiary branches, grain yield, and productivity. Means of two sets differed significantly for days to 50% flowering, flowering duration, plant height and pods per plant. Set-I had higher mean values for basal secondary branches, apical secondary branches, tertiary branches, and pods per plant, while Set-III had higher values for grain yield and productivity.

Variance Analysis

Residual (or restricted) maximum likelihood analysis for Set-I indicated that genotypic variance (σ^2_g) and genotype \times environment interaction variance (σ^2_{ge}) were highly significant for most traits across all environments. The Wald statistic was also highly significant indicating the adequacy of selected environments (E) for the evaluation. In Set-I, among vegetative traits, σ^2_g was highly significant for plant height and plant width (except in E1 and E2) in all seven environments, while σ^2_{ge} for basal and apical primary branches in E6, basal secondary branches in E4, apical secondary branches in E1, E4 and E6, and tertiary branches in E2 and E4 were highly significant. Genotypic variances for all the four reproductive traits (days to 50% flowering, flowering duration, days to maturity, and grain filling duration) were highly significant in all the environments. For yield

Table 3. Range and means of different traits pooled over environments in the large-seeded kabuli chickpea germplasm lines in three sets.

Trait	Set-I		Set-II		Set-III	
	Range	Mean	Range	Mean	Range	Mean
Days to 50% flowering	32.3–63.4	48.4	30.6–66.1	50.4	30.5–60.6	39.2
Flowering duration (days)	26.5–43.9	34.8	27.1–41.4	32.1	27.4–38.1	32.2
Days to maturity	97.7–118.7	111.8	95.6–124.4	115.7	90.2–116.8	103.7
Duration of grain filling (days)	50.8–73.2	62.8	57.0–77.1	65.7	53.0–72.0	64.6
Plant height (cm)	33.6–51.7	42.4	37.5–56.1	47.7	32.8–52.2	42.8
Plant width (cm)	40.7–59.2	50.8	36.2–51.8	45.6	31.6–42.0	37.6
No. basal primary branches	2.3–3.2	2.5	1.4–2.5	2.1	1.7–2.2	1.8
No. apical primary branches	1.7–3.0	2.1	0.8–2.4	1.6	1.4–2.8	1.8
No. basal secondary branches	1.6–2.6	2.4	0.6–2.2	1.3	0.3–1.8	0.7
No. apical secondary branches	2.9–7.7	4.6	1.6–7.9	3.4	0.6–6.6	2.3
No. tertiary branches	0.7–2.5	1.4	0.2–1.8	0.8	0.04–1.9	0.4
No. pods per plant	19.5–43.4	28.3	13.7–30.3	19.3	13.5–33.0	18.3
100-seed weight (g)	22.5–63.0	47.3	22.3–60.7	46.5	20.5–56.6	46.2
Grain yield (kg ha ⁻¹)	882–1430	1170	407–1544	1021	987–1856	1479
Per day productivity (kg ha ⁻¹ d ⁻¹)	6.6–13.6	10.6	3.1–13.7	8.9	8.6–17.6	14.5

and yield components, σ_{ge}^2 for pods per plant (except in E1 and E2) and 100-seed weight was highly significant in all environments, while σ_g^2 for grain yield and productivity were significant only in E2, E4, and E5 (data not given).

Residual (or restricted) maximum likelihood analysis of pooled data in Set-I indicated that genotypic variance (except basal primary, apical primary, and basal secondary branches) and σ_{ge}^2 (except basal primary and apical primary branches) were highly significant for all the vegetative traits (Table 4). For reproductive traits, both σ_g^2 and σ_{ge}^2 were highly significant whereas for grain yield components, though σ_g^2 was significant for all traits, σ_{ge}^2 was significant only for 100-seed weight (Table 4). Considering the high magnitude of σ_g^2 observed for most of the traits, it should be possible to select high-yielding genotypes with a combination of desirable traits such as early maturity, large seed size, and high yield. The significance of σ_{ge}^2 for 10 out of 15 traits studied indicates the extent of differential response of genotypes across environments. The high h_b^2 estimates (65–99%) for most of the traits indicate that the variation in this group of genotypes is highly heritable, even though the σ_{ge}^2 is highly significant but low in magnitude.

The Set-II was evaluated in five environments. Genotypic variance was significant for plant height and apical secondary branches among vegetative traits in all the five environments; plant width (except in E11), basal primary branches (except in E4), and tertiary branches (except in E9) in four environments; and apical primary branches (in E9 and E11) and basal secondary branches (in E8 and E11) in two environments. Except for flowering duration in E8, σ_g^2 for reproductive traits was highly significant in all the environments. Among yield related traits, 100-seed weight again showed highly significant σ_g^2 in all environments followed by grain yield and productivity in four environments (in E9). For pods per plant, the σ_g^2

was significant in only three environments (E4, E8, and E11) (data not given). The pooled analysis, except for pods per plant, revealed significant σ_g^2 and σ_{ge}^2 for all traits (Table 4). The estimates of h_b^2 were high for all the traits (65–98%) except for the number of basal primary, apical primary, basal secondary, and tertiary branches.

Residual (or restricted) maximum likelihood analysis in Set-III revealed significant σ_g^2 for most traits. Genotypic variance among vegetative traits was highly significant for plant height, apical secondary branches, and tertiary branches in all six environments; apical primary branches (in E9, E10, and E11) and basal secondary branches (in E1, E11, and E12) in three environments; plant width in E13; and basal primary branches in E10. Genotypic variance for the four reproductive traits (days to 50% flowering, flowering duration, grain filling duration, and days to maturity) was highly significant in all environments. In four of the six environments (E6, E10, E11, and E12), σ_g^2 for grain yield was significant, while for its associated components, such as 100-seed weight, the σ_g^2 was highly significant in all six environments, as in Set-I. The σ_g^2 was highly significant for pods per plant (E8, E9, E11, and E12) and productivity per day (E8, E10, E11, and E12) in four environments. Genotypic variance (except basal primary branches) and σ_{ge}^2 (except basal primary and secondary branches and pods per plant) in pooled analysis were significant for all the traits (Table 4). The h_b^2 values were high for all traits (67–99%). Overall, the Set-III genotypes also exhibited highly heritable genetic variability and the possibility of selecting superior genotypes.

The pooled analysis of all 18 trials in 13 environments showed highly significant σ_g^2 , σ_{ge}^2 , and residual (error) variance (σ_e^2) for all the traits (data not given). An overview of the variance analysis shows that plant height and width among vegetative traits varied greatly among genotypes in all sets with very high estimates of h_b^2 (82–96%). Apical secondary

Table 4. Genotypic (σ^2_g) and genotype \times environment (σ^2_{ge}) variances and broad-sense heritability (h^2_b) in multi-environment evaluation of three sets of large-seeded kabuli chickpea germplasm lines.

Trait	Set-I			Set-II			Set-III		
	σ^2_g	σ^2_{ge}	h^2_b	σ^2_g	σ^2_{ge}	h^2_b	σ^2_g	σ^2_{ge}	h^2_b
Days to 50% flowering	145.6**	7.4**	99.0	134.3**	12.5**	97.6	72.3**	16.6**	95.8
Flowering duration (days)	15.7*	30.9**	75.7	10.2**	13.0**	72.9	6.6*	8.4**	76.5
Days to maturity	41.0**	20.3**	90.9	72.1**	18.4**	93.9	39.3**	17.8**	94.2
Duration of grain filling (days)	47.4**	54.6**	84.8	24.7**	20.3**	81.2	21.6**	16.6**	88.2
Plant height (cm)	42.4**	4.9*	96.2	45.9**	6.2**	93.3	25.4**	6.1**	94.9
Plant width (cm)	47.2**	12.4**	94.1	24.5**	6.1**	88.0	6.3*	5.1**	81.8
No. basal primary branches	0.1	0.1	39.8	0.1**	0.1**	61.0	0.05	0.04	67.3
No. apical primary branches	0.1	0.5	32.5	0.1**	0.1*	62.0	0.1*	0.1*	77.6
No. basal secondary branches	0.2	0.5**	65.0	0.2**	0.2*	56.4	0.2**	0.05	82.7
No. apical secondary branches	1.3**	1.0**	85.1	1.0**	0.8**	76.6	2.1**	0.6**	92.5
No. tertiary branches	0.5**	0.2**	83.0	0.2*	0.3**	56.8	0.4**	0.3**	82.1
No. pods per plant	54.3**	8.1	90.1	27.4**	9.1	78.5	31.2**	3.3	92.3
100-seed weight (g)	139.1**	3.6**	99.2	77.3**	3.8**	98.1	100.4**	5.1**	98.7
Grain yield (kg ha ⁻¹)	62035**	9905	90.4	35941*	53288**	65.4	43362*	39821**	81.2
Per day productivity (kg ha ⁻¹ d ⁻¹)	6.6**	0.9	92.1	4.4**	5.0**	73.3	6.2**	3.9**	86.1

*Indicates significance at $p = 0.05$.

**Indicates significance at $p = 0.01$.

and tertiary branches exhibited highly significant σ^2_g and σ^2_{ge} in all sets, while σ^2_g and σ^2_{ge} for basal primary, apical primary, and basal secondary branches were significant only in Set-II. All the reproductive traits showed highly significant σ^2_g and σ^2_{ge} and high h^2_b in all sets (72–99%). The 100-seed weight among grain yield traits showed significant σ^2_g and σ^2_{ge} in all sets. Genotypic variance for pods per plant, grain yield, and productivity was significant in all the sets, while σ^2_{ge} was significant for grain yield and productivity in Set-II and Set-III. This implies that promising genotypes can be selected based on plant height, days to 50% flowering, days to maturity, and grain filling duration among vegetative traits. The number of pods per plant, 100-seed weight, and grain yield in all three sets also demonstrated highly significant and consistent σ^2_g coupled with very high estimates of h^2_b .

A review of the weather data in different seasons revealed no appreciable difference among environments for sunshine hours, minimum and maximum temperatures, and total pan evaporation during the cropping period. The major differences were in terms of irrigated or rainfed and the quantity of rainfall received during the cropping season (20 to 118 mm). In Set-I, a rainfall of 33 mm at podding stage during 2004–2005 increased the experimental mean yield over the previous year, both in irrigated and rainfed environments, by 2.8 and 40.2%, respectively. In Set-III, a rainfall of 26.6 mm at flowering stage during 2008–2009 increased the mean yield by 49.1% for irrigated and 66.4% for rainfed environments over the previous year. Regarding irrigated vs. rainfed environments, the mean yield decreased by 29 to 51% in rainfed compared to irrigated environments. These differences may not be entirely due to rainfall or irrigation alone and could be a consequence of substantial genotype \times environment interactions. Among genotypes, the yield reduction in

controls (small- and medium-seeded types) was high (35 to 46%), while in the selected large-seeded kabuli lines it varied from 9.1 (ICC 17450) to 38.7% (ICC 17109). On the other hand, the reduction in yield for genotypes ICC 17109, ICC 11883, and ICC 17457 was more than 30%. ICC 17450, an early, large-seeded genotype, showed only 9.1% reduction in yield under rainfed environment.

Principal Component Analysis

As σ^2_{ge} was highly significant for many traits, the PCA was performed on pooled values for different traits in each set separately for grouping the genotypes into separate clusters. The genotypes in each cluster that showed similar reaction to varying environments should possess the same common sensitive traits based on which of the PC axes cluster the genotypes. The first three PCs explained 81, 78, and 83% of the total variation respectively for Set-I, Set-II, and Set-III (data not given). The PC1 used six latent roots in all sets and PC2 used five latent roots in Set-I and Set-III and four in Set-II, while PC3 used only two latent roots in all three sets. Based on these latent roots, vector loadings were synthesized for each genotype (data not given) and the PCs separated the genotypes based on these vector loadings. Thus it appears that the PC1 differentiated genotypes based predominantly on vegetative and reproductive traits (40% variation explained), while PC2 used yield related traits (30% variation explained) in all sets.

Cluster Analysis

The grouping of all genotypes based on the scores of first three PCs resulted in three major clusters in Set-I (Fig. 1a), two clusters in Set-II (Fig. 1b), and three clusters in Set-III (Fig. 1c). All four control cultivars clustered together in

Set-I and Set-II, while in Set-III, L 550 (a high-yielding, small-seeded, late maturing widely adapted cultivar) clustered separately. Comparison of cluster means for various traits in all sets (Table 5) indicated that the clusters did not differ for branching traits, though in some cases the differences were significant due to low error variance. This indicates that there is little to choose from between clusters for these traits. All the clusters in all three sets differed significantly for flowering duration, days to maturity, grain filling duration, plant height and width, 100-seed weight, and productivity. Genotypes from complimentary clusters can be selected for recombinant breeding. For example, the genotypes in the second cluster of Set-I have significantly higher values for basal primary branches, pods per plant, grain yield, and productivity with low values for flowering duration, grain filling duration, and 100-seed weight; the genotypes in third cluster have exactly the opposite configuration. These genotypes are complimentary to each other and can be effectively used as parents in a breeding program.

Correlation Analysis

The correlation coefficients (Table 6) among all the 15 traits in three sets separately reveal the existence of significant and meaningful correlations (≥ 0.707 or ≤ -0.707 , $r^2 \geq 0.50$), between the following pairs of traits: days to 50% flowering and plant height, days to 50% flowering and plant width, days to 50% flowering and basal secondary branches, days to 50% flowering and apical secondary branches, days to 50% flowering and tertiary branches, flowering duration and grain filling duration, plant height and plant width, plant height and days to maturity, plant width and days to maturity, basal primary branches and basal secondary branches, basal secondary branches and tertiary branches, apical secondary branches and tertiary branches, and grain yield and productivity. Significant negative (≤ -0.707) correlations were observed between days to 50% flowering and grain filling duration, days to maturity and productivity, grain yield and 100-seed weight, and pods per plant and 100-seed weight pairs. The negative correlations between pods per plant and 100-seed weight, days to maturity and productivity, and grain yield and 100-seed weight needs to be considered when breeding for yield improvement. The negative correlation between days to maturity and productivity and pods per plant and 100-seed weight may be due to their simultaneous demand for photosynthates and other nutrients, as these traits are phenologically parallel in development. Improvements in the source capacity through improved canopy size and its efficiency for dry matter production, assimilation, and transport to the sink are prime factors that need to be considered for increasing yields in well-managed conditions. If the source capacity improves, the negative association between 100-seed weight and pods per plant can be broken, resulting in higher yields. However, increasing the canopy size is associated with delayed

flowering and late maturity, which may be a drawback for the rainfed environments where early maturity is desirable. An alternative strategy could be to decrease canopy size and increase the density of planting to maintain optimum number of pods per plant and maximize 100-seed weight. Thus the need for different plant types for purely rainfed and irrigated environments are imperative. In a previous study, it was reported that genotypes for favorable environments are different from those for marginal environments, though occasionally genotypes combining wider adaptability factors can be found (Arshad et al., 2003).

Selection could be based on both vegetative and reproductive traits in favorable environments but only reproductive traits such as yield per se and early maturity should be considered in unfavorable environments. Naidu et al. (1987) reported that selection based on yield would be most effective, followed by number of fruiting branches, when genotype \times environment interactions were significant. Generally, genotypes with low to medium 100-seed weight are high yielding, compared to genotypes with high 100-seed weight. However, high 100-seed weight lines are more stable in performance (Singh and Paroda, 1986). High 100-seed weight should be considered as one of the most important traits in kabuli types not only due to high price but because of its high stability and heritability. However, under rainfed conditions, high seed weight alone will not impart stability unless linked with fast grain filling, increased partitioning, and early maturity. Thus, simultaneous improvement of 100-seed weight and other traits like pods per plant and days to maturity is likely to result in increased yields.

Stability Analysis

Stability of genotypes for grain yield across environments was assessed using regression approach and deviation from regression. Finlay and Wilkinson (1963) proposed that linear response is positively associated with mean performance and that it may be considered as a measure of stability. However, Eberhart and Russell (1966) emphasized that both linear (b_i) and nonlinear (S^2d_i) components, in addition to mean, should be considered in judging the phenotypic stability of a particular genotype as their responses are independent from each other. In the present study, all genotypes showed a regression value of unity, except ICCV 2 in Set-I; ICC 14188 and ICC 14201 in Set-II; and ICC 17450, ICC 17456, and JGK 1 in Set-III, which showed significant deviation from unity (data not shown). The S^2d_i values ranged from nonsignificant to highly significant for the majority of the genotypes in all sets indicating that their performance cannot be predicted although most of them showed a regression value of unity. However, a few genotypes with high mean yield, regression values of unity, and very low S^2d_i values were selected from the three sets of materials tested in this study (Table 7).

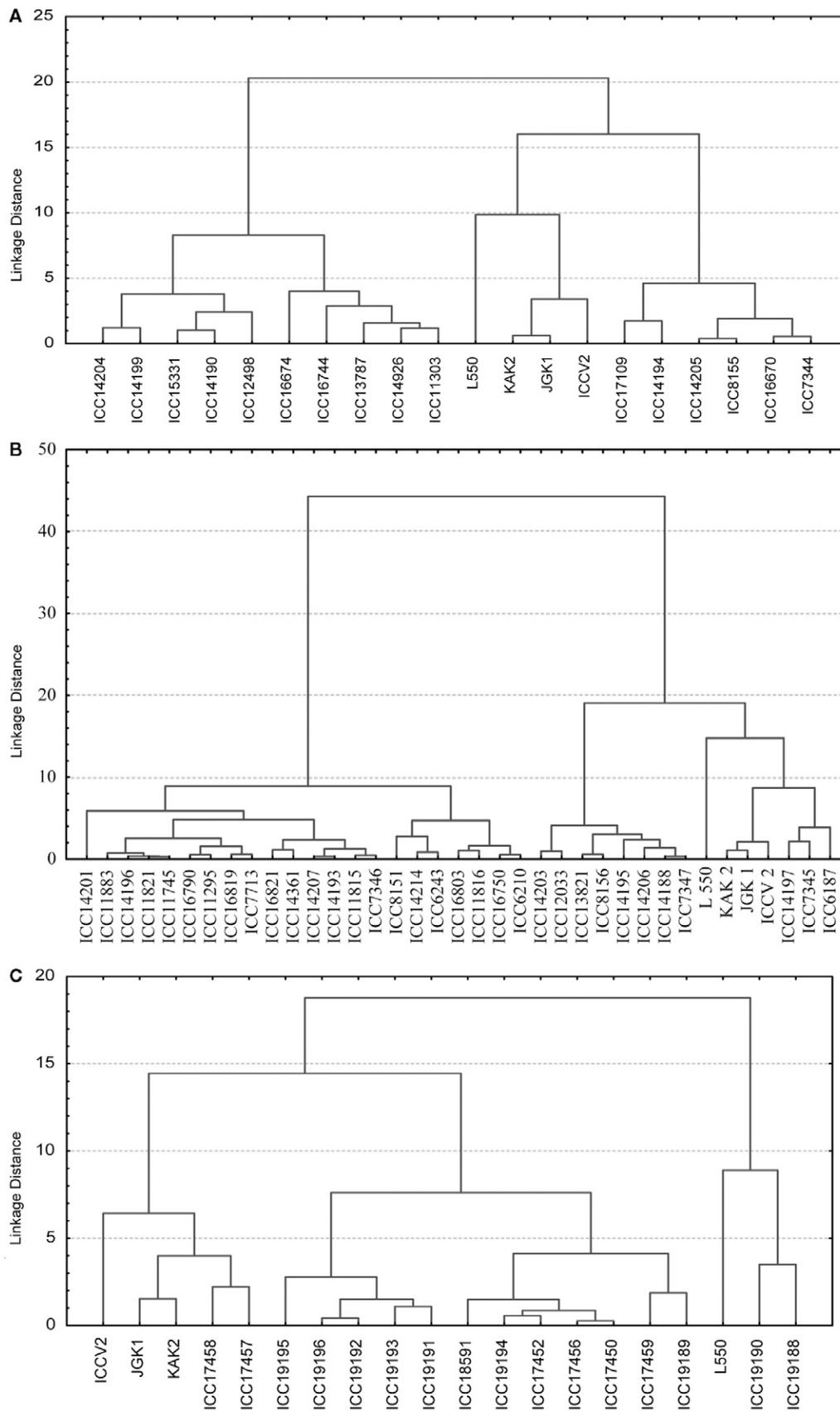


Figure 1a. Dendrogram of (a) 16 large-seeded kabuli chickpea germplasm lines of Set-I and control cultivars based on score of first three principal components (PCs), (b) 33 large-seeded kabuli chickpea germplasm lines of Set-II and control cultivars based on score of first three PCs, and (c) 16 large-seeded kabuli chickpea germplasm lines of Set-III and control cultivars based on score of first three principal components (PCs).

Table 5. Cluster means for different traits of three sets of large-seeded kabuli germplasm lines.

Trait	Cluster means [†]								
	Set-I			Set-II		Set-III			
	Cluster I	Cluster II	Cluster III	Cluster I	Cluster II	Cluster I	Cluster II	Cluster III	
Days to 50% flowering	57.1a	42.5b	36.4b	58.8a	38.0b	38.4b	35.2b	56.90a	
Flowering duration (days)	34.3b	30.0c	38.6a	31.0b	33.7a	29.5b	33.6a	30.9ab	
Days to maturity	116.4a	102.7c	107.8b	122.0a	107.2b	98.6b	102.7b	115.4a	
Duration of grain filling (days)	58.2b	59.6b	71.4a	63.3b	68.9a	60.2b	67.7a	58.7b	
Plant height (cm)	47.8a	34.8b	38.3b	52.5a	41.1b	38.8b	43.5ab	46.9a	
Plant width (cm)	56.0a	43.1b	45.6b	49.2a	40.6b	35.1b	37.9a	40.4a	
No. basal primary branches	2.6b	2.8a	2.5b	2.1a	1.9b	1.8b	1.8b	2.1a	
No. apical primary branches	2.4a	2.1a	2.1a	1.7a	1.5b	2.0a	1.8a	2.0a	
No. basal secondary branches	2.4a	2.8a	2.2a	1.5a	1.0b	0.6b	0.6b	1.5a	
No. apical secondary branches	4.9a	4.3a	3.8a	3.4a	3.3a	2.3b	1.7b	4.5a	
No. tertiary branches	1.6a	1.4a	1.0a	0.9a	0.6b	0.1b	0.3b	1.6a	
No. pods per plant	25.9b	41.2a	25.4b	18.6b	20.2a	21.5a	16.2a	21.3a	
100-seed weight (g)	48.3a	29.9b	56.4a	47.3a	45.3b	38.2b	51.7a	37.7b	
Grain yield (kg ha ⁻¹)	1166.6a	1405.5a	1086.6a	953.8b	1114.8a	1698.0a	1445.4b	1247.0b	
Per day productivity (kg ha ⁻¹ d ⁻¹)	10.1b	13.8a	10.1b	7.8c	10.5a	17.3a	14.1b	11.0c	

[†]Means followed by different letters are significantly different at $p = 0.05$.

The best genotypes in each set were compared for yield, yield components, and stability of performance across environments against two controls, L 550, and KAK 2. In Set-I, ICC 14190, a landrace from India, was ranked first with a mean yield of 1430 kg ha⁻¹. It has large 100-seed weight (37.4 g) and high productivity (13.64 kg ha⁻¹ d⁻¹) and is highly resistant to Fusarium wilt. The other two entries, ICC 14194 and ICC 7344, are early flowering, extra-large-seeded types with grain yields similar to the best control,

L 550 (Table 7). All these three genotypes exhibited high stability with regression value of unity and deviation near zero. The next entry, ICC 17109, is an extra-large-seeded type (63 g 100 seeds⁻¹) with a lower grain yield and low stability (highly significant S²d_v). In Set-II, ICC 7345, a breeding line from Mexico, yielded similarly (1352 kg ha⁻¹) to the best control, L 550 (1544 kg ha⁻¹). It is an early flowering line with relatively extra large seed size and high productivity. In Set-III, ICC 17457, a large-seeded, late flowering

Table 6. Phenotypic correlations between traits based on pooled mean in three sets of kabuli germplasm lines.

Trait combination	Correlation value		
	Set-I	Set-II	Set-III
Plant height and days to 50% flowering	0.833**	0.757**	–
Plant width and days to 50% flowering	0.851**	0.824**	0.515*
Days to maturity and days to 50% flowering	0.894**	0.784**	0.823**
Grain filling duration and days to 50% flowering	–0.705**	–0.877**	–0.664**
Basal secondary branches and days to 50% flowering	0.724**	–	0.666**
Apical secondary branches and days to 50% flowering	–	0.695**	0.755**
Tertiary branches and days to 50% flowering	0.514*	0.680**	0.769**
Grain filling duration and flowering duration	0.857**	0.632**	0.812**
Plant width and plant height	0.922**	0.952**	0.910**
Days to maturity and plant height	0.905**	0.924**	0.614**
Days to maturity and plant width	0.896**	0.926**	0.760**
Pods per plant and days to maturity	–0.695**	–0.611**	–0.772**
100-seed weight and grain filling duration	0.529*	0.475**	0.698**
Basal secondary branches and basal primary branches	0.670**	0.494**	0.783**
Apical secondary branches and basal secondary branches	0.750**	–	0.698**
Tertiary branches and basal secondary branches	0.680**	0.748**	0.904**
Tertiary branches and apical secondary branches	0.724**	0.938**	0.857**
100-seed weight and pods per plant	–0.857**	–0.823**	–0.916**
Grain yield and pods per plant	0.635**	0.588**	0.622**
Grain yield and 100-seed weight	–0.624**	–0.592**	–0.71**
Per day productivity and grain yield	0.947**	0.963**	0.961**

*Indicates significance at $p = 0.05$.

**Indicates significance at $p = 0.01$.

Table 7. Yield, yield components, and stability of the best large-seeded kabuli chickpea genotypes, across environments in three sets.

Genotype	Days to 50% flowering	Pods per plant	100-seed weight (g)	Grain yield (kg ha ⁻¹)	Productivity (kg ⁻¹ d ⁻¹)	b _i [†]	S ² d _i [‡]
Set-I							
14190 [§]	59.8	26.6	37.4	1430	13.64	0.70	6869
14194	37.1	31.2	51.3	1210	11.77	1.48	3638
7344	36.3	27.3	54.1	1169	10.74	0.76	3606
17109 [§]	35.0	23.6	63.0	1154	10.76	0.77	72420**
L 550	61.1	43.4	19.7	1413	12.90	1.44	-12867**
KAK 2	37.5	40.8	39.8	1294	15.23	1.12	-4994
LSD (5%)	2.9	8.2	3.2	271	2.53		
Set-II							
7345	35.0	19.3	48.2	1352	12.69	0.84	-7901
7347	41.1	16.1	50.7	1105	9.19	1.23	-6734
11883	59.4	18.1	60.7	890	7.16	0.83	75043**
L 550	60.9	40.6	19.1	1544	13.33	1.41	42854**
KAK 2	36.7	23.4	37.9	1263	12.57	0.90	152259**
LSD (5%)	2.5	6.0	2.4	233	2.12		
Set-III							
17457	49.9	15.0	49.5	1856	17.64	1.11	58536**
17452	35.7	17.6	54.0	1644	16.24	0.71	1456
19189	32.4	17.3	50.7	1549	15.66	0.69	-4726
19192	35.3	14.7	56.6	1312	12.70	0.85	2791
L 550	60.6	33.0	20.5	1671	14.93	1.18	338296**
KAK 2	35.6	19.0	37.7	1730	17.87	1.10	-16502**
LSD (5%)	1.6	5.0	2.3	207	2.08		

**Indicates significance at $p = 0.01$.

[†]b_i, linear component.

[‡]S²d_i, nonlinear component.

[§]Completely resistant to Fusarium wilt.

landrace from Mexico, had the highest mean grain yield (1856 kg ha⁻¹) but also had low stability (highly significant S²d_i). The other superior lines were ICC 17452 and ICC 19189, both early flowering, extra-large-seeded types with high stability. They were similar to the best control, KAK 2, for grain yield and days to 50% flowering but had higher 100-seed weight and greater stability.

Overall, large- and extra-large-seeded kabuli lines can be as high yielding as the best control and early in maturity. Results indicate that as the 100-seed weight increases, the pods per plant decreases, thus adversely affecting grain yield. The control cultivars showed highly significant S²d_i values and their yields varied widely across environments. Samuel et al. (1970) and Khan et al. (1987, 1988) suggested that the linear regression could simply be regarded as a measure of response of a particular genotype, which depends largely on a number of environments, whereas the deviation from regression line is a better measure of stability, and that genotypes with the lowest or nonsignificant deviations from regression are the most stable. Zubair et al. (2002) suggested that if regression coefficients of most of the genotypes do not significantly deviate from unity, the stability of these genotypes should be judged on two other parameters, that is, mean and S²d_i. Jain and Pandya (1988) reported that for any given situation the ideal genotype is one with high mean performance, linear response (b_i = 1),

and low nonlinear sensitivity coefficient (S²d_i = 0). These parameters help to select stable genotypes that interact less with the environment in which they are to be grown. Large amount of variability for seed weight, mainly represented by additive gene action, along with high heritability (Niknejad et al., 1971; Kumar and Singh, 1995) indicate that selection should be effective for significant improvement in seed size. In an earlier study, we found that seed size in chickpea was controlled by two genes that interacted through dominant epistasis and that a cross between small and normal seed size parents can give rise to a recombinant with better seed size (Upadhyaya et al., 2006c). But, as observed in the present study as well as in an earlier report (Upadhyaya et al., 2002), negative association of seed weight with other important traits such as pods per plant has to be considered when attempting to increase both seed weight and grain yield. Considering the association between important traits should enhance the ability to breed desirable genotypes using appropriate parents and selection procedures.

CONCLUSIONS

The 65 geographically diverse large-seeded chickpea genotypes evaluated in three sets exhibited large genetic variability for most traits studied except for branching traits, for which genotypic variance was minimal in all sets. Cluster analysis indicated that these lines were more diverse than

the control cultivars. The phenotypic correlations between branching traits and yield components were nonsignificant. The reproductive traits such as flowering duration, grain filling duration, and days to maturity did not show any significant correlation with branching traits except days to 50% flowering, which showed significant positive correlation with basal secondary, apical secondary, and tertiary branches, indicating that as chickpea has an indeterminate growth habit, branching continues parallel to extended vegetative growth phases including late flowering. The significance of σ_g^2 for plant height and plant width in all sets as well as their highly significant positive association with days to 50% flowering and days to maturity suggests that canopy size (plant height \times plant width) is positively correlated with crop duration. This confirms the prevalence of two different plant types, that is, early maturing and compact types for rainfed ecosystem and late maturing with large canopy types for intensive cultivation under irrigation. The compact types can be adapted for high density planting and complete their life cycle quickly to best utilize available resources such as soil moisture. They should show high harvest index, productivity, and high grain yield under post-rainy conditions in vertisols or under limited irrigation in alfisols with average management. On the other hand, under intensive management the crop is expected to produce a large canopy with more pod bearing branches and late flowering and maturity, with short flowering duration and long grain filling duration. In this case the vegetative sink and yield components do not compete with each other, as the plant would have completed its canopy structure by the time of flowering. Large leaf area would support a large sink composed of high pod number and large seed size, resulting in high yield. In both these plant types, sensitivity to environmental variation is expected to be minimum as the early maturing types escape stress and the better managed late maturing type is buffered against stress.

The promising genotypes identified in this study are large-seeded kabuli types with high yield and stable performance. These can be used in breeding program to develop large-seeded high-yielding kabuli cultivars or used directly for cultivation after evaluating their performance in large scale trials.

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