Multilocational Evaluation of Some Selected Chickpea Nodulation Variants in India

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Abstract

High- (HN) and low-nodulating (LN) selections from each of the two cultivars ICC 4948 and ICC 5003 developed at ICRISAT Asia Center, were evaluated in seven experiments in five different agroecological zones in India. The objectives of this multilocational experiment were to validate nodulation capacities of the selections in different environments, and to determine if the high-nodulating selections were indeed high yielding. Two nonnodulating selections were included as references to assess N_2 fixed by the different selections using the N difference method. Relative differences for nodule number and nodule mass between the HN and LN selections within a cultivar were consistent across locations and years. The HN selections generally yielded higher (range 4–41% in ICC 5003 HN and 4–106% in ICC 4948 HN) than the LN versions of the same cultivar, but the differences were significant ($P < 0.05$) only in two of the seven experiments.

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Estimates of nitrogen fixation, as measured by deducting the N-yield of nonnodulating lines from that of the nodulating lines, showed that HN lines had more fixed N in grains (2–44 kg N ha⁻¹) than those of LN selections. The long-duration nonnodulating chickpea line ICC 4993 NN grew well at Hisar and New Delhi (both 29° N), and should be preferred as the nonfixing control. At the other three locations, both ICC 4993 NN and ICC 4918 NN could be used as nonfixing control lines.

ICC 4948 HN was more susceptible to fusarium wilt than its parent, suggesting the need to ensure that selections are not inferior to parents for tolerance to diseases.

Introduction

In the process of biological nitrogen fixation (BNF) by legumes, both partners, the root nodule bacteria (*Rhizobium* or *Bradyrhizobium*) and the host plant have key roles. There are many constraints limiting the success of root nodule bacteria as inoculants. These include host, quality of inoculants, soil nitrogen (N) and phosphorus (P), high native rhizobial population, soil moisture, and interactions of these factors (Thies et al. 1991, Triplett and Sadowsky 1992). There is a considerable scope to enhance N_2 fixation by legumes through host-plant selection (Bliss 1985), in addition to selection of rhizobial strains for high N_2 fixation.

Variability in nodulation and N_2 fixation has been reported in different legumes such as faba bean, fieldpea, common bean, soybean, mungbean, cowpea, chickpea, and pigeonpea. Most work, however, has been on soybean, in which supernodulating lines have been selected (Carroll et al. 1985, Gremaud and Harper 1989, Francisco and Akao 1993) High-nodulating (HN) and low-nodulating (LN) lines have been reported in common bean (Pereira et al. 1993), chickpea (Rupela 1992, Rupela 1994), and pigeonpea (Rupela and Johansen 1995a, Dudeja and Khurana 1996). Intra-cultivaral variability for nodulation capacity in chickpea (*Cicer arietinum* L.) has recently been established in studies at ICRISAT Asia Center, where occurrence of nonnodulating, HN, and LN lines have been observed within several cultivars. All these nodulation selections were developed by pure line selection (Rupela 1992, Rupela 1994). Unlike the hypernodulating mutants of soybean, which were agronomically inferior to their normal nodulating parent cultivar (Wu and Harper 1991, Pracht et al. 1994, Song et al. 1995), the yield of HN selections was greater than those of the LN selections of the same cultivars in these studies (Rupela 1994). To study if the HN and LN selections of chickpea were stable for nodulation across locations, which are likely to differ in the population spectrum of native root-nodule bacteria and other soil and environmental factors, a multilocational experiment was conducted. Results of this experiment at five locations in India are described in this paper.

Materials and Methods

Chickpea, a cool-season legume is grown during the postrainy season (September/November to February/April) in India. It is generally grown on conserved soil moisture without irrigation. Also, rainfall during its growing period is occasional and scanty (Table 1). Seven experiments were conducted at five locations in India at research farms of Chaudhary Charan Singh Haryana Agricultural University (CCS HAU), Hisar, Haryana (29°N); Indian Agricultural Research Institute (IARI), New Delhi (29°N); Jawaharlal Nehru Krishi Vishwa Vidyalaya, Rafi Ahmad Kidwai (RAK) College, Sehore, Madhya Pradesh (27°N); Dr Panjabrao Deshmukh Krishi Vidyapeeth (PDKV), Akola, Maharashtra (23°N); and Marathwada Agricultural University, Agricultural Research Station (ARS), Badnapur, Maharashtra (20°N). Some soil characteristics of the experimental sites are given in Table 1. Five of the seven experiments (Table 1) were sown after a presowing irrigation provided to facilitate land preparation for sowing. One irrigation at flowering is a recommended practice at locations below 26°N or in light soils. The two experiments at Akola were irrigated thrice after sowing, an unusual practice.

The four test entries were HN and LN selections each from two chickpea cultivars ICC 4948 and ICC 5003. Their parent cultivar ICC 4948 (a low- to average-nodulating cultivar) and ICC 5003 (a high-nodulating cultivar), and two nonnodulating (NN) selections, one each from cultivars ICC 4993 and ICC 4918, were used as controls. The main plot had two N levels, low (N1) and high (N2), and the eight chickpea entries were the subplot treatments of split-plot design. No nitrogen was added to N1, while 100 kg N ha⁻¹ as urea was added to N2, 1 month before the sowing of chickpea, either to soil or to a previous cover crop sorghum. The subplot size was 4 × 3 m² at Hisar, Sehore, Badnapur, and 4 × 1.8 m² at Akola and New Delhi. The experiment had three replications at Hisar and four replications each at Akola, Badnapur, New Delhi, and Sehore. A basal dose of 17.5 kg P ha⁻¹ was applied at the time of sowing. Seeds of all the cultivars were uniformly inoculated with efficient *Rhizobium* strains (Table 1). ICC 4948 and its selections and ICC 4993 NN are long-duration lines, ICC 5003 and its selections are medium-duration lines, and ICC 4918 NN is a short-duration line.

Observations on nodule number, nodule, plant biomass, total dry matter, and grain yield were made at all the five locations (Table 1) on at least 10 plants per plot. Observations on acetylene reduction activity (ARA) were made only at IARI, New Delhi, on the same plants used for nodulation observations. Final yield of grain and stover were determined on a net plot size of 3.6 m² at Akola and New Delhi, and 7.2 m² at Hisar, Sehore, and Badnapur. A subsample from each plot was used to determine N concentration in the different cultivars to

Table 1. Characterization of sites at sowing and some agonomic and weather information, multilocational experiment at five locations in India, postrainy season, 1994/95, 1995/96.

Character	Akola 1994/95	Akola 1995/96	Badnapur 1995/96	New Delhi 1994/95	Hisar 1994/95	Hisar 1995/96	Sehore 1994/95
Soil characteristics at sowing							
Soil type	Vertisol	Vertisol	Vertisol	Sandy loam	Sandy loam	Sandy loam	Vertisol
pH (1:2.5)	8.1	8.1	8.0	8.5	7.8	7.6	7.6
Total N (%)	0.051	0.047	0.045	0.042	0.061	0.065	0.045–0.082
Mineral N ($\mu g\ g^{-1}$)	nd[1]	nd	nd	12	14–16	14–16	13–19
Organic matter (%)	nd	0.59	0.45	0.45	0.44	0.42	0.55
EC ($dS\ m^{-1}$)	0.24	0.23	0.30	1.0	0.30	0.29	0.34
Olsen-P ($\mu g\ g^{-1}$)	9.8	10.3	9.0	4.0	9.0	10.0	15.8
Sowing date	16 Nov 1994	4 Dec 1995	10 Nov 1995	30 Nov 1994	11 Nov 1994	23 Nov 1995	9 Nov 1994
Rhizobial strain for seed inoculation	Nil	AKCR 1	BCR72	SP 4	Ca 181	Ca 181	IC 59
Nodulation observation (days after sowing)	45	45	40	65	55	55	50
Final harvest (days after sowing)	112	113	124	175	164	150	133
Dates of postsowing Irrigation	22Nov1994 7 Dec 1994 5 Feb 1995	5 Dec 1995 18 Jan 1996 10 Feb 1996	10 Dec1995[2] 15 Feb 1996	Nil[2]	Nil[2]	Nil[2]	3 Jan 1995[2]
Rainfall (mm) between sowing and harvesting	24	9	Nil	63	85	9	75

1. nd = not determined.

2. One irrigation (presowing) was given, which was followed by land preparation and sowing, a common practice of the region.

calculate N uptake. N_2 fixed by the different cultivars was calculated by deducting N uptake by the nonnodulating lines from that by nodulating lines in a replication at the given N level (difference method).

Results and Discussion

Nodulation and Acetylene Reduction Assay

ICC 4948 HN generally formed significantly ($P < 0.05$) higher numbers of nodules than its parent ICC 4948, both at N1 and N2, except at Akola in the postrainy season 1995/96, and at Hisar at N2 in the postrainy season 1994/95 (Table 2). At Hisar, this selection was adversely affected by the soilborne fungal disease caused by *Fusarium oxysporum* f sp. *ciceri*. ICC 5003 HN nodulated similar to its parent ICC 5003 at all the locations except at Akola in 1994/95 and at N2 in 1995/96; at Badnapur in 1995/96, at Sehore in 1994/95 when it formed significantly ($P < 0.05$) greater number of nodules than its parent (Table 2).

Nodule mass was recorded near flowering at different ages [45–65 days after sowing (DAS), Table 1] of plants at different locations, and therefore, its comparison across locations is difficult. Still, most nodule mass was observed at Hisar in both the years (range 476–1336 mg $plant^{-1}$ at 55 DAS), and least nodule growth was observed at Badnapur (range 5–195 mg $plant^{-1}$) and in New Delhi (range 47–183 mg $plant^{-1}$, Table 3). As in case of nodule number per plant, HN selections of both the cultivars (ICC 4948 and ICC 5003) formed consistently high nodule mass at all the locations at both the soil N levels, except for ICC 5003 at N1 level at Akola in 1995/96, and for cultivar ICC 4948 at N2 level at Akola and Hisar in 1995/96. In five out of the seven experiments, the parent line ICC 4948 formed nodule mass closer to its LN selection (ICC 4948 LN). In the other two cases, it formed a nodule mass greater than its LN selection, but still lower than its HN selection. ICC 5003 is a high-nodulating cultivar, and ICC 5003 HN was selected to be only marginally superior to its parent. But ICC 5003 HN formed significantly ($P < 0.05$)greater nodule mass at N1 than its parent ICC 5003 in five out of seven experiments. The two nonnodulating lines ICC 4918 NN and ICC 4993 NN did not form any nodules in any of the seven experiments at the five locations.

Both nodule number and nodule mass are known to be suppressed due to high mineral N in soil (Rawsthorne et al. 1985, Rupela and Johansen 1995b, Streeter 1988). Mean (of all entries) nodule number per plant was lower at N2 than at N1 at all the five locations except in New Delhi, where 5% more nodules formed at N2. The extent of reduction in nodule number per plant ranged from 10% at Hisar in 1995/96 to 64% at Badnapur in 1995/96 (Table 2). Reduced nodule mass due to high soil N was recorded in four of the six experiments, where both low (N1) and

Table 2. Nodule number per plant of chickpea nodulation selections at five different locations in India, postrainy seasons, 1994/95 and 1995/96.

	Akola	Akola			Badnapur			New Delhi			Hisar						Sehore		
		1995/96			1995/96			1994/95			1994/95			1995/96			1994/95		
Chickpea line	1994/95	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean
ICC 4948 HN	41	29	27	28	61	52	56	37	33	35	16	21	19	50	40	45	20	16	18
ICC 4948 LN	11	14	15	14	21	6	14	9	9	9	11	8	9	10	11	11	10	9	9
ICC 4948	19	24	27	26	28	13	21	16	9	13	9	17	13	17	14	16	16	12	14
ICC 5003 HN	36	30	29	29	82	19	51	31	38	35	22	20	21	35	31	33	21	14	18
ICC 5003 LN	21	25	18	22	15	3	9	9	15	12	9	10	10	9	24	17	11	9	10
ICC 5003	26	34	20	27	59	6	33	30	40	35	38	25	32	36	27	31	15	12	14
ICC 4993 NN	0[1]	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
ICC 4918 NN	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
SE	±4.0	±4.19	(4.01)[2]	±2.9	±0.6	(0.6)	±0.1	±4.9	(5.2)	±3.7	±3.7	(3.3)	±2.3	±2.6	(2.4)	±1.8	±0.2	(0.3)	±0.2
Mean	nr[3]	20	17		33	12		17	18		13	13		20	18		12	9	
SE	nr	±2.2			±0.3			±1.0			±2.2			±1.1			±0.1		

1. Chickpea lines without nodules were excluded from statistical analysis.
2. Values in parentheses are SEs to compare means at the same level of N.
3. nr = not relevant.

Table 3. Nodule dry mass (mg $plant^{-1}$) of chickpea nodulation selections at different locations in India, postrainy seasons, 1994/95 and 1995/96.

	Akola	Akola			Badnapur			New Delhi			Hisar						Sehore		
		1995/96			1995/96			1994/95			1994/95			1995/96			1994/95		
Chickpea line	1994/95	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean
ICC 4948 HN	511	258	254	384	96	70	83	105	106	106	872	442	657	930	583	757	235	204	220
ICC 4948 LN	373	202	219	292	53	5	29	75	62	69	386	106	246	573	476	525	126	111	119
ICC 4948	386	212	242	307	89	16	53	90	63	77	299	429	364	907	527	717	167	148	158
ICC 5003 HN	509	264	317	400	195	43	19	128	183	156	845	352	599	1336	733	1035	328	237	283
ICC 5003 LN	279	277	233	252	59	2	31	47	49	48	126	158	142	726	546	637	143	120	132
ICC 5003	366	242	317	323	147	4	76	157	155	156	687	305	496	836	600	718	266	212	239
ICC 4993 NN	0[1]	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
ICC 4918 NN	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
SE	±40.3	±17.1	(16.8)[2]	±12.1	±0.1	(0.1)	±0.01	±13.9	(14.3)	±10.1	±57.8	(63.3)	±44.7	±74.8	(55.6)	±39.3	±2.7	(2.7)	±1.9
Mean	nr[3]	176	197		80	17		75	77		402	224		604	433		158	129	
SE	nr	±8.8			±3.2			±4.8			±2.1			±61.6			±1.2		

1. Chickpea lines without nodules were excluded from statistical analysis.
2. Values in parentheses are SEs to compare means at the same level of N.
3. nr = not relevant.

high (N2) levels were included. The extent of reduction in the four experiments ranged from 18% at Sehore in 1994/95 to 79% at Badnapur in 1995/96. At the other two locations, a 3% increased nodule mass was recorded in New Delhi and 12% at Akola. Even at these two locations, a small reduction (2–30%) in nodule mass was recorded in two of the six nodulating lines. Interactions between genotypes and N-levels for nodule number and nodule mass per plant were statistically significant in three experiments and nonsignificant in the other three experiments (Tables 2 and 3).

Acetylene reduction assay (ARA) to compare N_2 fixation between and among the different lines was done only at IARI, New Delhi. Acetylene reduction assay values (µmoles C_2H_4 produced per plant per hour) of ICC 5003 HN were significantly superior to those of ICC 5003 LN, both at N1 and N2. There were no consistent trends for ICC 4948 HN (Table 4). Acetylene reduction assay is a point-in-time observation, and has been criticized for its validity (Minchin et al. 1994). It may, however, detect large treatment differences, for example, between ICC 4948 HN and ICC 4948 LN (Rupela et al. 1995). But this was not so in this case and this is surprising, particularly when nodulation differences between HN and LN selections were large (Table 3).

Table 4. Acetylene reduction assay of chickpea nodulation selections at the Indian Agricultural Research Institute, New Delhi, postrainy season 1994/95.

	Acetylene reduction assay					
	µM C_2H_4 plant^{-1}h^{-1}			µ M C_2H_4 g^{-1} nodule mass		
Chickpea line	N1	N2	Mean	N1	N2	Mean
ICC 4948 HN	4.8	4.3	4.6	46	42	44
ICC 4948 LN	3.4	4.4	3.8	44	62	53
ICC 4948	4.9	1.9	3.4	53	29	41
ICC 5003 HN	5.4	10.8	8.1	47	59	53
ICC 5003 LN	1.6	2.6	2.1	29	51	40
ICC 5003	6.2	3.8	5.0	38	24	31
ICC 4993 NN	0	0	0	0	0	0
ICC 4918 NN	0	0	0	0	0	0
SE	±1.10(1.10)[1]		±0.76	±7.9 (8.2)		±5.8
Mean	3.3	3.5		32	33	
SE	±0.38			±1.6		

1. Values in the parentheses are SEs to compare means at the same level of N.

Total Dry Matter and Grain Yield

Only grain yield was assessed at final harvest at Akola in 1994/95 and at Badnapur in 1995/96. In the other five experiments where both total dry matter and grain yield were measured (Tables 5 and 6), Hisar was the highest-yielding location in both 1994/95 and 1995/96 seasons, followed by New Delhi. Generally, short-duration lines perform best at lower latitudes and long-duration lines perform well at higher latitudes (Saxena 1987). The nonnodulating lines ICC 4993 NN and ICC 4918 NN yielded the lowest in all the seven experiments, except at Hisar in 1994/95. ICC 4918 NN, a short-duration line, yielded low even at high soil N conditions (Tables 5 and 6) at Hisar and New Delhi. At the other three locations (Akola, Badnapur, Sehore), the yields of ICC 4993 NN and ICC 4918 NN at N2 level were similar. This suggests that both cultivars can be used as non N_2-fixing references at Akola, Badnapur, and Sehore (all <25°N latitude), while at Hisar and New Delhi (both 29°N latitude), ICC 4993 NN, a long-duration line, should be preferred. The yield of ICC 4993 NN was exceptionally high at Hisar in 1994/95, suggesting high soil-N status at the experimental site. Its high yield, close to or better than that of the HN line ICC 5003 (a medium-duration line) could also be due to its duration being longer than that of ICC 5003.

The yield differences among the six nodulating lines were generally nonsignificant in most experiments. The HN selection ICC 5003 HN produced higher grain yield than ICC 5003 LN in all the seven experiments at the five locations, and the increase ranged from 4 to 41%. In six of the seven experiments, it also yielded higher than its parent ICC 5003 (Table 6). The beneficial effect of higher nodulation of ICC 5003 HN than that of its parent ICC 5003 was not marked in case of total dry matter (Table 5), but was apparent for grain yield (Table 6). This perhaps suggests a better partitioning of its resources into grains by ICC 5003 HN. The HN selection ICC 4948 HN yielded higher than ICC 4948 LN at all the locations except at Hisar and at N2 level in New Delhi. The increase in yield ranged from 4 to 106% at the different N levels and locations. At Hisar, a significant number of plants of ICC 4948 HN died due to fusarium wilt, which apparently affected its yielding capacity. This was an important observation, suggesting that care should be exercised in selection for high N_2-fixing lines such that the selected material should not be more susceptible to stress factors than their parents.

The selection ICC 4948 HN yielded 5 to 42% higher than its parent in four of the five experiments (data from Hisar not considered due to the problem of fusarium wilt). ICC 5003 HN yielded 2 to 29% higher than its parent ICC 5003 in all the seven experiments, and the two N levels.

Super-nodulating mutants of soybean (Eskew et al. 1989; Wu and Harper 1991) and *Phaseolus* bean (Buttery et al. 1990) are known to produce significantly lower yield than their normal nodulating parents. In contrast, the HN selections reported

Table 5. Total dry matter (t ha^{-1}) of chickpea nodulation selections at four different locations in India, postrainy seasons, 1994/95 and 1995/96.

	Akola			New Delhi			Hisar						Sehore		
	1995/96			1994/95			1994/95			1995/96			1994/95		
Chickpea line	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean
ICC 4948 HN	2.82	3.69	3.26	5.83	4.74	5.29	6.00	5.97	5.56	5.83	4.58	5.21	2.55	3.31	2.93
ICC 4948 LN	2.10	2.28	2.14	5.57	4.95	5.26	9.72	9.58	9.65	6.39	6.46	6.42	2.06	2.76	2.41
ICC 4948	2.15	2.75	2.45	6.04	4.64	5.34	8.89	9.17	9.03	6.45	6.66	6.56	2.56	3.16	2.86
ICC 5003 HN	2.57	3.39	2.98	6.33	5.94	6.13	7.78	6.53	7.15	3.85	4.58	4.22	2.85	3.49	3.17
ICC 5003 LN	2.23	3.26	2.75	5.73	6.38	6.06	7.92	6.39	7.15	4.27	3.89	3.96	2.74	3.42	3.08
ICC 5003	2.03	2.85	2.44	5.39	5.42	5.40	7.78	6.81	7.29	4.38	4.46	4.26	2.99	3.41	3.20
ICC 4993 NN	1.52	2.29	1.90	5.47	5.47	5.47	7.50	8.89	8.19	1.81	2.74	2.27	1.98	2.75	2.37
ICC 4918 NN	1.74	2.26	2.00	4.09	3.85	3.97	2.78	1.85	2.31	2.50	3.06	2.78	1.65	2.44	2.04
SE	±0.183 (0.191)[1]		±0.135	±0.467 (0.469)		±0.332	±0.525 (0.523)		±0.37	±0.192 (0.189)		±0.136	±0.117 (0.108)		±0.076
Mean	2.13		2.85	5.56		5.17	7.29		6.79	4.41		4.51	2.42		3.09
SE		±0.0.37			±0.159			±0.192			±0.083			±0.06	

1. Values in parentheses are SEs to compare means at the same level of N.

Table 6. Grain yield (t ha^{-1}) of chickpea nodulation selections at different locations in India, postrainy seasons, 1994/95 and 1995/96.

	Akola				Badnapur			New Delhi			Hisar						Sehore		
		1995/96			1995/96			1994/95			1994/95			1995/96			1994/95		
Chickpea line	1994/95	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean
ICC 4948 HN	1.50	1.59	1.85	1.61	0.96	1.00	0.98	1.32	1.05	1.20	1.44	1.35	1.40	1.95	1.02	1.49	1.30	1.58	1.44
ICC 4948 LN	0.73	1.06	1.09	0.90	0.75	0.71	0.73	1.27	1.37	1.32	2.50	3.13	2.81	2.50	1.69	2.09	1.04	1.34	1.19
ICC 4948	1.06	1.24	1.53	1.22	0.83	0.82	0.82	1.34	1.23	1.28	2.57	2.70	2.63	1.23	1.51	1.37	1.23	1.51	1.37
ICC 5003 HN	1.30	1.34	1.78	1.43	1.08	1.18	1.13	1.64	1.59	1.62	2.12	1.74	1.93	1.59	1.17	1.38	1.50	1.80	1.65
ICC 5003 LN	1.17	0.94	1.56	1.21	0.95	0.94	0.95	1.49	1.54	1.51	1.68	1.49	1.68	1.43	1.17	1.30	1.44	1.73	1.59
ICC 5003	1.14	1.16	1.59	1.25	1.05	1.07	1.06	1.27	1.45	1.36	1.74	1.70	1.72	1.58	1.17	1.38	1.46	1.77	1.61
ICC 4993 NN	0.66	0.60	0.98	0.71	0.78	0.78	0.78	1.22	0.96	1.09	2.23	2.54	2.41	0.66	0.92	0.79	0.69	1.06	0.87
ICC 4918 NN	0.49	0.81	0.92	0.68	0.69	0.99	0.84	0.28	0.38	0.33	0.25	0.14	0.19	0.52	1.15	0.84	0.87	1.29	1.08
SE	±0.015	±0.195 (0.201)[1]		±0.138	±0.061 (0.057)		±0.041	±0.176 (0.184)		±0.130	±0.176 (0.184)		±0.130	±0.111 (0.123)		±0.079	±0.057 (0.052)		±0.037
Mean	nr[2]	1.10	1.42		0.90	0.93		1.85	1.85		1.65	1.85		1.54	1.21		1.19	1.51	
SE	nr		±0.039			±0.028			±0.037			±0.037			±0.048			±0.029	

1. Values in parentheses are SEs to compare means at the same level of N.
2. nr = not relevant.

here generally produced yields at least marginally superior to those of their parents.

High soil N level (N2) resulted in 7% reduced total dry matter (means of all six nodulating cultivars) at two of the five locations (New Delhi in 1994/95 and Hisar in 1994/95) where it was measured (Table 5). However, such a reduction was not recorded for grain yield in these two experiments (Table 6). This could be due to interactions for partitioning of resources into grains and/or the problem of fusarium wilt at Hisar. A 21% reduced grain yield at N2 over N1 was recorded at Hisar in 1995/96. At the other locations, a 21–34% increase was recorded for total dry matter, and a 3–29% increase in grain yield at N2 over that at N1. Reduction in grain yield due to N application has also been reported by Rupela et al. (1997) and its reason(s) need to be studied.

Nitrogen Fixation by Difference Method

Grain N concentration was determined only at Akola, Badnapur, and Sehore, and stover N concentration only at Akola in 1995/96. Therefore, the quantity of fixed N in stover could be determined only in the Akola experiment in 1994/95 (data not shown) and that in grain for the four experiments at the three locations (Table 7). At all the three locations, both N1 and N2 grain of ICC 4948 HN had more (8.4–43.3 kg N ha^{-1}) fixed N than did ICC 4948 LN and its parent ICC 4948. Similarly, grain of ICC 5003 HN had more (2.3–12.6 kg N ha^{-1}) fixed N both at N1 and N2 in all the four experiments (where it was measured), than ICC 5003 LN. Also, ICC 5003 HN had more (1.6–9.5 kg N ha^{-1}) fixed N in grain than its parent except at N2 at Akola in 1995/96 when it was lower by 1.4 kg N ha^{-1} (Table 7). In most cases, the superiority of the two HN selections was statistically nonsignificant.

The amount of N_2 fixed by the different chickpea lines in the four experiments are small (1.2–50 kg N ha^{-1}) and therefore the additional N_2 fixed (2.3–43.3 kg N ha^{-1}) due to the higher nodulation capacity of the HN selections. In high producing environments the level of N_2 fixation is likely to be high and thus the benefits of HN selections are also likely to be higher. This needs to be examined. There is also a need to compare the difference method with a more reliable method such as ^{15}N enrichment and natural abundance.

Conclusions

The LN and HN selections from the two cultivars nodulated consistently across locations (Tables 2, 3), and there were significant differences ($P < 0.05$) in their nodulation capacities at most locations. Any small increase in yield due to HN selections should be welcomed because the HN selections generally yielded more and fixed more N (even if marginally) than LN selections and their parents (Tables 6 and 7).

Table 7. Fixed N (kg ha^{-1}) in grains of chickpea nodulation selections measured by difference method at five different locations in India, postrainy seasons 1994/95 and 1995/96.

	Akola				Badnapur			Sehore		
		1995/96			1995/96			1994/95		
Chickpea line	1994/95	N1	N2	Mean	N1	N2	Mean	N1	N2	Mean
ICC 4948 HN	54.3	36.8	40.1	35.8	11.6	8.3	10.0	22.5	22.6	22.6
ICC 4948 LN	11.0	20.5	26.9	23.7	1.5	0[1]	0	9.4	11.7	10.5
ICC 4948	25.2	27.6	31.7	29.7	1.2	0	0	18.1	18.9	18.5
ICC 5003 HN	32.4	32.5	38.5	35.5	17.6	16.8	17.2	32.2	33.4	32.8
ICC 5003 LN	21.6	22.6	36.2	29.4	8.0	4.2	6.1	25.3	28.9	27.1
ICC 5003	31.1	25.1	39.9	32.5	11.1	7.3	9.2	28.6	31.8	30.2
SE	±2.14	±5.02(5.49)[2]		±3.88	±2.68 (2.48)		±1.75	±2.41 (2.05)		±1.45
Mean	29	27.5	35.5		8.5	4.1		22.7	24.6	
SE	nr[3]		±0.31			±1.45			±1.52	

1. Negative values were calculated during analysis which were obviously artifacts and perhaps due to relatively unadapted nature of the selections than that of the nonnodulating reference of similar duration. These were replaced by zero.

2. Values in parentheses are SEs to compare means at the same level of N.

3. nr = not relevant.

The presence of HN plants (higher than the mean nodulation of the parent) seems to be a widely occurring phenomenon (Rupela and Johansen 1995b), and the search for more versions of these can be extended to agronomically accepted, diseases- and other stress-tolerant lines. In multilocational experiments, characterization of the experimental sites should include determination of mineral N at sowing. Physiological parameters, e.g., days to 50% flowering and maturity affecting yield of cultivars should also be recorded. The methods used for characterization of soil parameters should be the same across locations. Most participating locations in this report did not have ready access to soil analysis facilities for variables reported in Table 1, and the soil samples had to wait for long periods before analyses, or were not done. Data on N_2 fixation quantification should be assembled in all the experiments. It seems that $\delta^{15}N$ analysis can be reliable only from some sites (Peoples, Turner, et al., these proceedings). Any future experiments should be restricted only to such sites. Most importantly, the high N_2-fixing selections must be at par (if not better) with their parents for the degree of tolerance to stress factor(s).

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