International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru, Andhra Pradesh 502 324, India

## SCREENING OF CICER SPECIES AND CHICKPEA GENOTYPES FOR RESISTANCE TO MELOIDOGYNE JAVANICA $^{1}$

by S. B. Sharma, Onkar Singh, R. P. S. Pundir and D. McDonald

**Summary**. Twenty-five accessions of *Cicer bijugum, C. chorassanicum, C. cuneatum, C. judaicum, C. pinnatifidum*, and *C. reticulatum*, 173 chickpea advanced breeding lines and five cultivars were evaluated for resistance to *Meloidogyne javanica* in a greenhouse. Resistance evaluation was on a 1-9 damage index (1=highly resistant; 9=highly susceptible) based on gall number, gall size, and galled area of root. Numbers of egg sacs were also counted. None of the tested lines was free of nematode damage. Damage indices of *Cicer* spp. ranged between 5.2 and 8.8. ICCW 47, an accession of *C. cuneatum* was less susceptible than other accessions. Variation in gall size was greater than that for gall number and galled area. Most of the tested lines showed symptoms of stress in terms of premature drying of leaves, chlorosis, and stunting of plants. Plant growth of two breeding lines (ICCV 90043 and 90243) and a cultivar (ICCC 42) was not affected by nematode parasitism, foliage of these genotypes remaining dark green and without premature leaf drop.

Chickpea (Cicer arietinum L.) is a host for the root-knot nematodes, Meloidogyne incognita (Kofoid et White) Chitw., M. javanica (Treub) Chitw., and M. artiellia Franklin (Nene et al., 1989) which cause substantial yield losses (Sikora and Greco, 1990; Sharma and McDonald, 1990). There are few management options available to resource-poor farmers to reduce damage caused to chickpea by root-knot nematodes. There are no commercial chickpea cultivars with resistance to Meloidogyne spp. but resistance to M. artiellia and Heterodera ciceri has been found in wild Cicer spp. (Di Vito et al., 1988; Greco, pers comm., Istituto di Nematologia Agraria, Italy). This paper describes the reactions of 25 accessions of six wild Cicer spp., 173 advanced breeding lines, and five chickpea cultivars to M. javanica.

## Materials and methods

Seeds of 14 accessions of *Cicer judaicum* Boiss., 2 of *C. bijugum* K. H. Rech., 4 of *C. pinnatifidum* Jaub. *et* Spach, 3 of *C. reticulatum* Ladiz, and 1 each of *C. cuneatum* Hochst. *ex* Rich and *C. chorassanicum* (Bge) M. Pop. were obtained from the Genetic Resources Unit of the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT),

Patancheru, Andhra Pradesh, India. Four seeds of each accession were sown in four 15-cm diameter pots containing a 3:1 (V:V) mixture of autoclaved riverbed sand and black cotton soil. Eggs of M. javanica were extracted from 8-week-old cultures maintained on tomato (Lycopersicon esculentum Mill. cv. Rutgers) by treating the roots with 0.1% sodium hypochlorite (Hussey and Barker, 1973). Ten to fifteen thousand eggs were added to the soil along with the seeds (2500-3750 eggs per seed). Eight weeks after seedling emergence, plants were carefully removed from the pots and roots evaluated for nematode damage in terms of gall number, gall size, and percentage of galled area of root, based on visual assessment of root area covered by galls to the total root area. Nematode reproduction was measured by counting the numbers of egg sacs after treating the roots with 0.25% trypan blue for 3-5 minutes. The egg sacs were stained deep blue, whereas roots were not stained.

Roots of each plant were evaluated for number of galls, size of galls, percentage of galled area of root, and egg sacs using a 1 to 9 index. Gall index (GI): 1 = 0 galls, 2 = 1-5, 3 = 6-10, 4 = 11-20, 5 = 21-30, 6 = 31-50, 7 = 51-70, 8 = 71-100, 9 = > 100 galls. Gall size (GS): 1 = no galls, 3 = very small, 5 = small, 7 = medium, and 9 = big galls. Percentage galled area (GA): 1 = no galls, 3 = 1-10% root area galled, 5 = 11-30%, 7 = 31-50%, and 9 = > 50% root area galled. A

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damage index (DI) was calculated as GI+GS+GA/3. Accessions with DI = 1.0 were considered highly resistant, DI  $\leq$  3.0 as resistant, DI  $\leq$  5.0 as moderately resistant, DI  $\leq$  7.0 as susceptible, and DI  $\leq$  9.0 as a highly susceptible.

Chickpea cultivar K 850 was used as highly susceptible check in all the tests. Data on GI, GS, GA and DI were statistically analysed and average indices are shown in Table I. DI was used to evaluate the reaction of chickpea genotypes

TABLE I - Reaction of accessions of wild Cicer spp. to Meloidogyne javanica.

ICCW No. <sup>1</sup>	Origin	Gall index	Gall size	galled area %	Damage index
		Cicer juda	icum		
73	Syria	8.6 <u>a</u>	5.7 <sup>c-f</sup>	$7.0^{a-d}$	7.1
74	Syria	9.0 <sup>a</sup>	5.7 <sup>c-f</sup>	8.3 <sup>abc</sup>	7.7
, i 75	Syria	8.7 <sup>a</sup>	6.1 <sup>b-f</sup>	8.1 <sup>abc</sup>	7.6
76	Syria	8.0 <sup>a</sup>	5.0 <sup>def</sup>	6.5 <sup>a-d</sup>	6.5
77	Syria	$9.0^{a}$	5.5 <sup>c-f</sup>	8.1 <sup>abc</sup>	7.5
78	Ethiopia	8.6 <sup>a</sup>	7.0 <sup>bc</sup>	6.6 <sup>a-d</sup>	7.4
79	Ethiopia	8.1 <sup>ab</sup>	6.1 <sup>b-f</sup>	6.1 <sup>bcd</sup>	6.8
80	Syria	7.5 <sup>abc</sup>	$4.8^{ m efg}$	6.8 <sup>a-d</sup>	6.4
81	Syria	9.0 <sup>a</sup>	5.0 <sup>d-g</sup>	8.3 <sup>abc</sup>	7.4
82	Syria	7.8 <sup>ab</sup>	5.0 <sup>d-g</sup>	7.4 <sup>a-d</sup>	6.5
83	India	$8.8^{a}$	7.0 <sup>bc</sup>	$8.8^{a}$	8.2
84	India	$9.0^{a}$	5.7 <sup>c-f</sup>	7.7 <sup>a-d</sup>	7.5
89	India	$9.0^{2}$	6.4 <sup>b-d</sup>	7.9 <sup>abc</sup>	7.8
90	India	8.8ª	5.6 <sup>c-f</sup>	8.2 <sup>abc</sup>	7.5
		C. bijugu1	n		
71	Syria	9.0 <sup>a</sup>	$9.0^{a}$	8.4 <sup>ab</sup>	8.8
72	Syria	8.8 <sup>a</sup>	$9.0^{a}$	7.8 <sup>abc</sup>	8.7
		C. pinnat	ifidum		
11	Syria	$8.0^{a}$	5.7 <sup>c-f</sup>	7.3 <sup>a-d</sup>	7.0
85	Syria	6.6 <sup>bc</sup>	4.6e-h	7.8abc	6.3
86	Syria	6.6 <sup>bc</sup>	4.8 <sup>cfg</sup>	$7.4^{a-d}$	6.3
88	Syria	$9.0^{a}$	5.0 <sup>d-g</sup>	9.0 <sup>a</sup>	7.5
		C. reticule	atum		
8	Syria	$8.4^{a}$	7.6 <sup>ab</sup>	7.0 <sup>a-d</sup>	7.7
48	Syria	8.1 <sup>ab</sup>	7.6 <sup>ab</sup>	7.6 <sup>ab</sup>	7.8
49	Syria	8.0 <sup>ab</sup>	5.0 <sup>d-g</sup>	8.4 <sup>ab</sup>	7.1
		C. cuneat	um		
47	Ethiopia	6.6 <sup>bc</sup>	3.0 <sup>in</sup>	6.0 <sup>cd</sup>	5.2
		C. choras	sanicum		
26	Afghanistan	9.0 <sup>a</sup>	5.0 <sup>d-g</sup>	6.3 <sup>bcd</sup>	6.8

 $<sup>^{1}</sup>$  ICRISAT chickpea wild species accession number. Data flanked on the columns by the same letters are not statistically different for P = 0.05, according to Duncan's multiple range test.

and accessions of *Cicer* spp. because gall number, gall size and galled area of root are intrinsic components of damage caused by root-knot nematodes. Numbers of egg sacs were rated using the scale developed for gall number.

The accessions were tested in a greenhouse between November 1989 and May 1990 (maximum temperature 22 - 32 °C, and minimum temperature 20 - 23 °C). The pots were irrigated regularly. Arnon's nutrient solution and 250 ppm nitrogen as ammonium nitrate were added to the pots every week (Arnon, 1938).

Seeds of advanced breeding lines and promising cultivars were received from the ICRISAT Chickpea Breeding Unit. The breeding lines were ICCVs 13, 14, 88102, 88103, 88106, 88109, 88110, 88405, 89230, 89244, 89307, 89314, 89344, 89402, 89445, 89701, 89702, 90001-90054, 90101-90152, 90202-90236, and 90240-90253. The promising cultivars developed at ICRISAT were ICCCs 37, 40, 42, ICCVs 2, and 5. All these genotypes were screened between July 1990 and December 1991 for resistance to *M. javanica* by calculating the DI based on GI, GS and GA as described above. Symptoms of stress on the foliage due to nematode parasitism on roots were visually assessed and genotypes with no apparent stress symptoms were identified.

## Results and discussion

All the accessions of Cicer spp. were susceptible to M. javanica (Table I). Root galling was extensive in most entries. Average GI ranged from 6.6 on ICCWs 47, 85, and 86 to 9.0 on ICCWs 26, 71, 74, 77, 79, 81, 84 and 88. Average GS was greatest on accessions of C. bijugum. Average GS of C. judaicum accessions ranged from 4.8 (ICCW 80) to 7.0 (ICCWs 78 and 83). Accessions of C. pinnatifidum and C. chorassanicum had small galls and C. cuneatum had very small galls. Variation in gall size was greater than in gall number and galled area of root in the Cicer spp. GA ranged between 6.0 (ICCW 47) and 9.0 (ICCW 88). DI of all the accessions except ICCW 47 was greater than 6. Large numbers of egg sacs (ratings 7-9) were observed on all the accessions. Almost all the genotypes showed symptoms of stress in terms of premature drying of leaves, leaf drop, chlorosis, bronzing of leaves, and stunting of the plants. The symptoms were similar to phosphorus and sulphur deficiencies in chickpea.

All the tested breeding lines were highly susceptible to *M. javanica*, and more than 25% were killed. The latter had very severe infection of root knot and 75% root area was galled, and over 100 egg sacs were present. Yellowing of leaves was the most common symptom. ICCV 90043, 90243 and ICCC 42 had DI greater than 6 but plant growth

was apparently not affected, and foliage was dark green and leaves did not drop as in other lines.

Di Vito et al. (1988) found high levels of resistance to cyst nematode, H. ciceri, in accessions of C. bijugum. Resistance to M. artiellia is available in C. judaicum, C. pinnatifidum, C. chorassanicum and C. cuneatum. It is apparent that wild Cicer spp. do not have resistance to M. javanica. Cicer cuneatum (ICCW 47) appears to be 'less susceptible' than accessions of other species and evaluation of more accessions of this species is desirable. Elite breeding lines and cultivars developed at ICRISAT Center lack resistance to M. javanica; however, reactions of ICCV 90043, 90243 and ICCC 42 require further testing preferably in the nematode infested fields. Resistance to M. incognita and M. javanica is generally based on number of galls and/or egg sacs (Hasan, 1983, Handa et al., 1985, Sharma et al., 1988, Mishra and Gaur, 1989). For comprehensive assessment of damage, use of the damage index. and egg sac index is suggested.

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## Literature cited

- Arnon D. I., 1938. Macro elements in culture solution experiments with higher plants. *Amer. J. Bot.*, 25: 322-325.
- DI VITO M., GRECO N., SINGH K. B. and SAXENA M. C., 1988. Response of chickpea germplasm lines to *Heterodera ciceri* attack. *Nematol. medit.*, 16: 17-18.
- HANDA D. K., JAIN K. K., MISHRA A. and BHATNAGAR S. M., 1985. Screening chickpeas for resistance to the root-knot nematode, Meloidogyne javanica. Intl. Chickpea Newsl., 12: 33.
- HASAN A., 1983. Reaction of chickpea cultivars to root-knot nematode. Intl. Chickpea Newsl., 8: 26-27.
- Hussey R. S. and Barker K. R., 1973. A comparison of methods of collecting inocula for *Meloidogyne* spp., including a new technique. *Pl. Dis. Reptr.*, 57: 1025-1028.
- MISHRA S. D. and GAUR H. S., 1989. Reaction of some chickpea, Cicer arietinum L., germplasm to the root-knot nematode, Meloidogyne incognita. Int. Nematol. Network Newsl., 6: 21-23.
- Nene Y. L., Shiela V. K. and Sharma S. B., 1989. A world list of chickpea (*Cicer arietinum* L.) and pigeonpea (*Cajanus cajan* (L.) Millsp.) pathogens. Legumes Pathology Progress Report-7. Patancheru, Andhra Pradesh 502 324, India: International Crops Research Institute for the Semi-Arid Tropics. 23 pp.
- SHARMA G. L., MATHUR B. N. and TAGI R. N. S., 1988. Screening of chickpea cultivars for resistance to the root-knot nematode, *Meloidogyne incognita. Intl. Chickpea Newsl.*, 18: 12.
- SHARMA S. B. and McDonald D., 1990. Global status of nematode problems of groundnut, pigeonpea, chickpea, sorghum and pearl millet and suggestions for future work. *Crop Protection*, 9: 453-458.
- Sikora R. A. and Greco N., 1990. Nematode parasites of food legumes. *In Plant-parasitic nematodes in Subtropical and Tropical Agriculture*, 181-235 (Eds M. Luc, R. A. Sikora and J. Bridge). Wallingford: CAB International.

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