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Mechanism of resistance to *Heterodera cajani* in *Cajanus platycarpus* accessions

Zubaida Elyas and Shashi B. Sharma

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

Abstract. Invasion of second-stage juveniles of *Heterodera cajani* was much quicker in the roots of susceptible *Cajanus cajan* genotype ICPL 87 than in the susceptible and resistant genotypes of *C. platycarpus*. Within 48 h, 16 juveniles were in the roots of ICPL 87 and less than 3 in the roots of ICPW 543, ICPW 544, ICPW 545 and ICPW 63. The nematode invasion continued to be greater in the roots of ICPL 87 than in the roots of other genotypes. In ICPW 63 (susceptible check) the nematode increased sharply after three days of inoculation. The three resistant genotypes differed in their response and nematode invasion in the roots of ICPW 543 within 48 h was twice as much as in the roots of ICPW 544 and ICPW 545. Pre- and post-infectional factors affecting resistance are discussed.

Keywords. *Cajanus platycarpus, Heterodera cajani,* mechanism of resistance, pigeonpea.

INTRODUCTION

Pigeonpea cyst nematode (*Heterodera cajani*) is an important pest of pigeonpea (*Cajanus cajan*) in India. The nematode was first reported on pigeonpea in 1967 (Koshy, 1967) and during the last 30 years it has been reported from all the major pigeonpea growing regions in India (Sharma and Sharma, 1993; Sharma *et al.*, 1992). The nematode causes significant damage to pigeonpea particularly on the black cotton soils (Vertisols) in the southern and western India. Efforts have been made at the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) Asia Center to identify sources of resistance to this nematode species in pigeonpea genepool. Evaluation of more than 7000 accessions of pigeonpea germplasm revealed that resistance to *H. cajani* is not readily available as most of the tested germplasm lines were susceptible or highly susceptible. Screening of wild relatives of pigeonpea, however, revealed that resistance to the cyst nematode is available in three accessions (ICPW 543, ICPW 544 and ICPW 545) of *Cajanus platycarpus* (Saxena *et al.*, 1996; Sharma, 1995). The nematode resistance in these accessions was purified (Sharma, 1995) and seeds collected after several cycles of purification were deposited in the pigeonpea genebank at ICRISAT. The accession numbers of the nematode resistant purified germplasm lines are ICPW 543, ICPW 544, and ICPW 545.

The objective of this investigation was to understand the mechanism of nematode resistance in the three accessions of *C. platycarpus*.

MATERIALS AND METHODS

Seeds of *Heterodera cajani* resistant *Cajanus platycarpus* ICPW 543, ICPW 544, and ICPW 545 and *H. cajani* susceptible ICPW 63 and ICPL 87 were obtained from the Genetic Resources Division of ICRISAT. ICPL 87 is a high yielding nematode susceptible pigeonpea cultivar. All the seeds of *C. platycarpus* accessions were mechanically scarified to facilitate germination.

Nematode population

The population of *Heterodera cajani*, collected from a pigeonpea field at the research farm of ICRISAT, was increased on pigeonpea cultivar ICPL 87. The nematode eggsacs and young cysts were collected from roots of 6-week old seedlings. The eggsacs and cysts were incubated at room temperature for extraction of second-stage juveniles. The juveniles were stored at 10°C for less than 48 h before inoculation.

Nematode invasion and development

Autoclaved sand+Vertisol (silty clay loam, 39% sand, 20% silt and 41% clay, pH 8.0) mixture (3:1, v/v) was filled in 80 7.5-cm-diam plastic pots. For each *C. platycarpus* accession and ICPL 87, there were 20 pots each sown with pregerminated seeds. In each pot, 1000 second-stage juveniles were inoculated near the roots of the seedling. The pots were irrigated daily and nutrient solution was added to the soil every five days. At 2, 4, 7, 9, 11, 14, 16, and 18 days of germination,

roots of two seedlings for each of the five accessions were stained in 0.05% cotton blue lactophenol for 2-3 min. The roots were spread on 22.0 cm×9.5 cm glass plates and number of juveniles in the roots was determined by observation with a stereoscopic microscope. The rate of nematode invasion was assessed by counting the juveniles in the roots under the stereoscopic microscope, and growth and development of the invaded juveniles was studied under high resolution Olympus BH 2 microscope.

RESULTS AND DISCUSSION

Invasion of second-stage juveniles of *H. cajani* was much quicker in the roots of susceptible *C. cajan* genotype ICPL 87 than in the susceptible and resistant genotypes of *C. platycarpus*. Within 48 h, 16 juveniles were in the roots of ICPL 87 and 0-3 in the roots of ICPW 543, ICPW 544, ICPW 545 and ICPW 63. The nematode invasion continued to be greater in the roots of ICPL 87 than in the roots of other genotypes. In ICPW 63 (susceptible check) the nematode increased sharply after three days of inoculation. The three resistant genotypes differed in their response and nematode invasion in the roots of ICPW 543 within 48 h was twice as much as in the roots of ICPW 544 and ICPW 545. However, there was a marked difference in the development of juveniles in roots of these genotypes. There was no nematode in roots of ICPW 543 after 7 days, and in the roots of ICPW 544 after 11 days. The invaded juveniles did not enter into their third stage on ICPW 543, and did not develop after fourth stage on ICPW 544. Males were observed after 8 days of inoculations on ICPW 545 and after 10 days on the susceptible genotypes, ICPL 87 and ICPW 63. Nematode infection on the Rhizobium nodule was more frequent on ICPW 545 than on other genotypes. After 20 days of inoculations, no females or cysts were observed on ICPW 543 and 544, two cysts were formed on ICPW 545 and 34 cysts with eggsacs on ICPW 63.

These results indicated that the *H. cajani* resistance in the *C. platycarpus* accessions was a combination of the pre-infectional as well as post-infectional factors. The pre-infectional resistance appeared to be milder in ICPW 543 than in the ICPW 544 and ICPW 545. The development of juveniles in roots was arrested within 7 days of root invasion in ICPW 543 whereas in ICPW 544 at least one juvenile reached up to the fourth stage. ICPW 545 apparently had a different mechanism of resisting the nematode development than that in ICPW 543 and ICPW 544 and apparent tendency of the juveniles was to develop into males. The nematode had a very reduced level of reproduction on ICPW 545.

Cai *et al.* (1997) reported that the gene which conferred resistance to the beet cyst nematode (*Heterodera schachtii*) encoded an LRR-containing protein. The encoded protein led to developmental arrest of the nematode and breakdown of the feeding structure. Similar mechanism of resistance to *H. cajani* is envisaged in the two accessions of *C. platycarpus*. As genes for resistance to the cyst nematode are not readily available, the resistance in *C. platycarpus* accessions can be transferred to pigeonpea cultivars. The wild species of *Cajanus*, which are closely related with *C. cajan*, represent a secondary gene pool and have been utilized to produce interspecific derivatives (Remanandan, 1990; Saxena *et al.*, 1987), whereas wild species that are not crossable with pigeonpea represent the tertiary genepool (Maeson, 1990). *Cajanus platycarpus* belongs to the tertiary genepool and the barriers to hybridization are post-zygotic, and now techniques are available to overcome these barriers (Mallikarjuna and Moss, 1995).

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