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International Journal of Current Research
Vol. 5, Issue, 09, pp.2459-2461, September, 2013

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

ISSN: 0975-833X

RESEARCH ARTICLE

REACTION OF COWPEA VARIETIES TO *Meloidogyne incognita* INFESTATION IN GHANA

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ARTICLE INFO

Article History:
Received 22nd June, 2013
Received in revised form
19th July, 2013
Accepted 25th August, 2013
Published online 14th September, 2013

Key words:

Meloidogyne incognita,
Resistance, Susceptibility,
Vigna unguiculata.

ABSTRACT

Ten cowpea varieties were preliminarily evaluated for their reaction to *M. incognita* under screen house condition in a Randomised Complete Block design with three replications. Three week old seedlings were inoculated with 350 and 700 second stage juveniles and set on concrete benches. Based on the reproduction factor and root gall index, Asontem and Asetenapa were considered to be resistant. Tona, Nhyira, Padituya, Hewale, Asomdwe and IT89KD-347-57 were considered susceptible whilst Adom and Videza were found to be tolerant. At the highest inocula level of 700J2/pot, Videza recorded an average yield of 6.4g which was significantly different from 2.3g as recorded for Hewale. The highest pod number of 8.0 was significantly different from 3.7 recorded for Videza and Asontem, considered to be tolerant and resistant at the highest inocula level respectively. Adom and Asetenapa recorded the highest and lowest plant height of 74.7cm and 19.3cm respectively at 700J2/pot. With the exception of Hewale, all the susceptible varieties recorded low yields at higher level of inoculation compared to the lower level. Based on the results of the study, Asontem and Asetenapa were identified as resistant to *M. incognita*.

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INTRODUCTION

Cowpea, *Vigna unguiculata* (L.), Walp is the most useful legume in West Africa (Langyintuo *et al*., 2000). It is an important food legume and essential component of cropping systems in the drier regions of the tropics and subtropics (Singh *et al*., 2003). In Ghana, it is one of the widely cultivated legumes mainly in the savanna and transitional zones (CRI, 2006). It's inclusion in the traditional farming systems is ideal as it supports sustainable land use by improving soil fertility and serving as rotation crop. However, plant parasitic nematodes most especially *Meloidogyne incognita* (Kofoid et White) constitutes a major constraint to cowpea production in most growing areas of the world (Sikora *et al.*, 2005). An estimated 59% loss of cowpea grain yield caused by *M. incognita* was reported in Nigeria (Ogunfowora, 1976), while Olowe (1978) found a seedling mortality rate of up to 80% for cowpea grown in soil with *M. incognita* population density of 1300 second-stage juveniles/L. Various control options are employed for the management of root knot nematodes but the resistance-strategy appears the most feasible especially for the low key peasant farmers who constitute majority of the farming population. Several works to identify resistance in cowpea have recorded some success for the management of root knot nematodes in various countries (Adegbite *et al*., 2005; Ehler *et al*., 2000, Fassuliots, 1979). In Ghana, the use of resistant cultivars, as a control option, has not been given an adequate attention especially with cowpea production and there is also a scarcity of published information. The present study, therefore, was aimed at evaluating the reaction of 10 cowpea varieties to different levels of *M. incognita* under plant house conditions.

MATERIALS AND METHODS

Experimental site

The trial was conducted at the Plant house of the Plant Pathology Section of the Council for Scientific and Industrial Research-Crops Research Institute (CSIR-CRI), Kumasi, Ghana from June to September, 2012.

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Source of cowpea varieties

Cowpea varieties, Tona, Nhyira, Adom, Asetenapa, Padituya, Asontem, Hewale, IT89KD-347-57, Videza and Asomdwe from the Legume and Oil Seed Improvement Division of the CSIR-CRI were used for this study.

Nematodes extraction and estimation

Root knot nematodes were collected from roots of infested tomato plants from farmers' fields at Asante Akim Agogo in the Ashanti region of Ghana. *M. incognita* was identified as described by CIH (1978). It was then maintained on Pectomec, a local tomato cultivar acquired from local farmers. Eight weeks after inoculation, heavily galled roots from tomato roots were cut into 1 cm long pieces, and eggs extracted by shaking for 3 minutes in a 0.5% solution of sodium hypochlorite (Hussey and Barker, 1973), in a glass jar. The eggs were then sieved through a 75 µm sieve nested onto a 38 µm pore sized sieve. The eggs on the 38 µm sieve were rinsed with tap water to reduce the concentration of the 0.5% sodium hypochlorite and collected in a 2-litre glass beaker. The egg-water suspension was mixed and diluted with tap water. The egg-water suspension was allowed to stand on laboratory benches under room temperature for the eggs to hatch into second stage juveniles. One ml aliquot, in three replicates, was taken with a pipette to estimate total number of the second stage juveniles in the suspension under stereoscopic microscope. The volume of the suspension in the beaker was then concentrated so that 1ml of the suspension contained 100 second stage juveniles.

Sowing and inoculation of cowpea varieties

Cylindrical plastic pots of 10cm radius were filled with steam sterilized sandy loam soil. Three seeds of each cowpea varieties was planted in each pot. One week after planting, the seedlings were thinned out to one seedling per pot. Three weeks after planting, the seedlings were inoculated with different levels of *M. incognita* second stage juveniles. This was done by carefully exposing the roots of the seedlings and with the aid of a graduated pipette the juveniles were collected and the roots inoculated with the infective second stage juveniles as per the inoculum levels. There were three replications at three levels (Pi: 0,350, and 700 J2/plant). The inoculated seedlings were place on concrete platforms and arranged in randomized complete block design. Watering was done when required.

Harvesting and Data collection

Fifty days after inoculation, the pods were harvested and the plant uprooted from the soil. Plant height was measured weekly after the seedlings were inoculated. Gall index, J2/ 200g soil, J2/ 5g roots, reproduction factor and yield data were taken at harvest. Roots of the cowpea plants were assessed for presence of galls and scored on a scale of 0-10 as described by Bridge and Page (1980). Host reaction of the cowpea cultivars and line were determined and ranked into resistance (GI<2, RF<1), tolerant (GI≤2, RF>1), hyper-susceptible (GI> 2, RF< 1) and susceptible (GI> 2, RF > 1) as described by Canto-Saez (1983).

Statistical analysis

Nematode count data was square root [$\sqrt{(x+0.5)}$] transformed to comply with the assumption of normal distribution. Data for the two planting times were pooled and analysed together using Genstat 8.1 (Lawes Agricultural Trust, VSN International) statistical package. Least significant difference (Lsd) at 5% was used for comparing mean differences.

RESULTS

Results from the study showed that the cowpea crops reacted differently to the different levels of the inocula. At an inocula level of 350 J2/pot, second stage juveniles extracted from 5g root of cowpea ranged from 14.7 to 1226.3 for Asomdwe and IT89KD-347-57 respectively (Table 1). Whilst at the highest level of 700J2/pot Nhyira recorded the highest number of 1642.0 second stage juveniles compared with 64.3 juveniles for Asetenapa (Table 1). At both inocula levels, it was observed that there was a significant difference in the number of juveniles extracted from the resistant and susceptible varieties. At inocula level of 700J2/pot, 2093.0 and 19.3 juveniles were recovered from the soil of Nhyira and Asontem respectively. At 350J2/pot, juveniles recovered from the soil of susceptible cultivar were different except for Nhyira and Hewale (Table 1). With the exception of Hewale, Tona and Padituya which showed no difference in juveniles recovered from the soil at inocula level of 700J2/pot, all the other susceptible varieties showed difference. At both inocula levels, (350J2/pot and 700J2/pot), there was a significant difference between juveniles recovered from the soils of the susceptible and resistant varieties (Table 1). Considering the nematode reproduction factor and galling index, the cowpea varieties were ranked as resistant (GI <2, RF<1), tolerant (GI≤2, RF>1), hyper-susceptible (GI> 2, RF< 1) and susceptible (GI> 2, RF > 1). Six varieties, Asomdwe, Tona, Nhyira, Padituya, IT89KD-347-57 and Hewale were susceptible to *M. incognita* (Table 1). For these gemplasm, the reproduction factor ranging from 1.3 to 2.9 and galling index ranging from 1.6 to 2.9 were significantly higher than Asontem and Asetenapa which were found to be resistant (Table 1). The reproduction factor for the

Table 1. Reaction of cowpea varieties to different inoculum levels of *M. incognita*

varieties	J2/5g root		J2/200g soil		Galling index (0-10)		Rep factor		Host efficiency
	350	700	350	700	350	700	350	700	
Tona	323.3 (17.9)	413.0 (20.3)	95.7 (9.8)	750.0 (27.4)	7.3 (2.8)	4.0 (2.1)	1.4 (1.3)	1.8 (1.5)	Susceptible
Nhyira	189.3 (13.7)	1642.0 (40.5)	597.7 (24.5)	2093.0 (45.6)	4.7 (2.3)	7.0 (2.7)	1.2 (1.3)	3.7 (1.9)	Susceptible
Hewale	427.7 (20.4)	674.3 (25.9)	553.3 (23.5)	836.0 (28.9)	6.0 (2.6)	8.0 (2.9)	3.0 (1.9)	2.3 (1.7)	Susceptible
IT89KD-347-57	1226.3 (36.4)	1107.3 (33.3)	1510.0 (38.9)	1610.0 (40.2)	6.3 (2.6)	7.7 (2.9)	8.4 (2.9)	3.8 (2.1)	Susceptible
Asetenapa	24.7 (4.9)	64.3 (8.0)	19.3 (11.3)	120.7 (11.0)	2.0 (1.6)	2.0 (1.6)	0.3 (0.8)	0.3 (0.8)	Resistant
Videza	268.3 (16.3)	311.0 (17.6)	1.0 (0.71)	21.0 (4.3)	2.0 (1.6)	2.0 (1.9)	1.0 (1.1)	0.4 (1.1)	Tolerant
Adom	190.0 (13.7)	200.0 (14.1)	95.3 (9.8)	111.3 (10.0)	2.0 (1.6)	2.0 (1.6)	0.9 (1.2)	0.8 (1.1)	Tolerant
Asomdwe	14.7 (3.9)	70.0 (8.3)	42.3 (6.4)	46.7 (6.8)	5.0 (2.4)	5.3 (2.4)	0.9 (1.2)	0.9 (1.2)	Susceptible
Padituya	141.7 (11.9)	880.0 (29.6)	390.0 (19.6)	872.0 (29.6)	7.0 (2.7)	7.7 (2.9)	2.0 (1.5)	2.8 (1.8)	Susceptible
Asontem	70.7 (8.4)	129.0 (11.4)	16.7 (4.4)	19.3 (4.8)	2.7 (1.8)	3.0 (1.9)	0.3 (0.9)	0.8 (0.9)	Resistant
Lsd	3.3	(2.3)	(4.4)	(3.3)	(0.9)	(0.6)	(0.1)	(0.3)	
CV	13.0	(6.3)	(11.8)	(9.1)	(5.0)	(6.6)	(1.7)	(13.5)	

Table 2. Effect of different inoculum levels on plant height, number of pods and yield of cowpea varieties

	Effect of different inocula levels on								
	Height (cm)			Number of pods			Yield (g)		
Varieties	0J2/pot	350J2/pot	700J2/ pot	0J2/pot	350J2/pot	700J2/Pot	0J2/pot	350J2/Pot	700J2/Pot
Tona	21.2	20.8	20.3	8.3	3.3	7.3	6.3	5.7	4.2
Nhyira	23.2	20.5	25.5	8.0	6.3	7.7	5.9	5.3	4.0
Hewale	20.8	23.1	23.7	8.3	4.3	4.7	4.0	1.6	2.3
IT89KD-347-57	36.3	36.7	40.7	6.0	4.7	4.7	4.0	3.4	2.4
Asetenapa	22.0	27.3	19.3	10.0	8.7	7.0	2.6	2.4	3.9
Videza	22.0	23.2	21.3	8.0	10.7	8.0	9.0	6.7	6.4
Adom	76.7	31.0	74.7	8.3	6.0	7.7	5.7	5.5	4.7
Asomdwe	30.8	33.2	26.3	3.7	4.3	4.0	3.4	3.1	2.6
Padituya	32.0	25.8	22.0	4.7	5.3	5.3	4.4	4.4	3.3
Asontem	63.1	47.3	54.5	4.3	6.0	3.7	5.6	4.1	5.6
Lsd (5%)	22.2	19.3	25.10	3.1	2.1	2.5	1.3	1.6	1.6
CV (%)	37.2	39.0	44.6	6.1	20.2	24.3	14.5	21.8	24.3

tolerant cultivars were not significantly different from those of the resistant varieties (Table 1). Plant height at 350J2/pot ranged from 20.8cm to 47.3cm for Tona and Asontem respectively. Whilst at 700J2/pot it ranged from 20.8cm to 74.7cm for Tona and Adom respectively (Table 2). There was a significant difference in plant height between Adom and all the other cowpea varieties except for Asontem at an inoculum level of 700J2/pot. Number of pods ranged from 3.3 to 13.0 for Tona and Videza respectively (Table 2). There was a significant difference in the mean number of pods between Videza and all the other varieties at 350J2/pot. At 700J2/pot, there was difference in pod number between two susceptible varieties Nhyira and IT93K-129-4 and two resistant varieties Asontem and Asetenapa (Table 2). Mean seed yield ranged from 1.6g to 9.0g for 350J2/pot and 0J2/pot respectively (Table 2). At 700J2/pot, mean yield for Videza and Asontem was found to be significantly higher than all the varieties considered to be susceptible; Tona, Padituya, Nhyira, IT89KD-347-57 and Hewale (Table 2).

DISCUSSION

The cowpeas evaluated showed a varied reaction from each other in their response to *M. incognita* infestation. The differences in reaction might be due to genetic variability among the varieties (Khan and Khan, 1991). Two of the cultivars, Asontem and Asetenapa were found to be resistant. Resistance to nematode infestation can be either pre-infection resistance, where the nematodes cannot enter the roots of the plant due to the presence of toxic chemicals or post-infection resistance in which nematodes are able to penetrate roots but fail to develop (Bandezu and Starr, 2003). The poor reproduction and galling of the resistant varieties suggest that resistance may be mediated before entry of the nematode through root exudates (Williamson and Hussey, 1996). The resistive nature of the cultivars may also be due to the formation of lignin, apposition of callose or wound periderm plant responses to limit the invasion of the nematode. The high parasitisation of Asomdwe, Tona, Nhyira, Padituya, IT89KD-347-57 and Hewale by the nematode indicates the weakness in their defense mechanism. This shows that they lack the genes to stop the penetration, development and reproduction of the nematode. It may also be due to the low level of glycosides in the plant tissue on which the enzyme glycosidase from the nematodes may act upon to liberate adequate free phenol for suppression of the nematode reproduction and development (Olowe, 2009). Adom and Videza, the tolerant varieties, although yielded satisfactorily is detrimental to subsequent crops as it could leave a high a population density of the nematodes in the soil. The differences in plant height and yield might depend on the different reaction of the crop to the nematode. Increase in plant height according to Pattison (2007) is due to the reduction in the number of galls and decrease in the activities of the nematodes. The reduction in activities of the nematode leads to a higher uptake of nutrients by the roots which subsequently improve the yield. Resistant cultivars have been found to increase and stabilize yield in a similar manner to treating high yielding susceptible cultivars with nematicides. Growing resistant cultivars may also increase the yield by 19-69% or even up to five times that of highly susceptible cultivars (Kinloch *et al.*, 1988; Young and Hartwig, 1988).

Conclusion

Asontem and Asetenapa can further be evaluated under field conditions to confirm their degree of resistance and further release for use by farmers in managing *M incognita* in Ghana.

Acknowledgement

The authors are grateful to the AGRA-Cowpea project of the CSIR-Crops Research Institute for sponsoring this work.

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