

## EFFECT OF *MELOIDOGYNE GRAMINICOLA* ON ROOT GROWTH OF RICE AND *ECHINOCHLOA COLONUM* L.

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### Abstract

The effect of *Meloidogyne graminicola* on root morphology and growth of rice and the *Echinochloa colonum* grass was investigated under glasshouse conditions. Invasion by nematode juveniles reduced total root length and the length of axes and laterals in both hosts, while the number of axes (nodal) in infested rice roots increased due to nematode invasion. On infested roots of both hosts, the laterals grew in clusters and appeared earlier than on uninfested roots. When a root was invaded by the nematodes and a gall formed, the growth of the root was checked and secondary laterals grew on the gall. Upto 11.6% in rice and 27.3% of total laterals in *E. colonum* were growing on the actual galls formed due to nematode invasion.

**Key words:** root-knot nematode, *Meloidogyne graminicola*, *Echinochloa colonum*, *Oryza sativa*, rice, root growth, root morphology.

### Introduction

Root-knot nematode *Meloidogyne* species may impair plant growth by inhibiting new root development, suppressing the rate of root extension and causing degeneration of existing roots (Hussey, 1985; Shane & Barker, 1986). The rice root-knot nematode *Meloidogyne graminicola* damages root growth by delaying new root development, suppressing the rate of root extension and altering the root growth pattern (Soomro & Hague, 1992a, 1992b). This abnormality in root growth leads to reductions in root length and nutrient uptake.

This study was conducted to compare the plant growth in general and root growth in particular of rice (*Oryza sativa* L.) and its weed *Echinochloa colonum* L., infested with a common nematode parasite, *Meloidogyne graminicola* Golden & Birchfield 1965.

### Materials and Methods

Seedlings of rice (cv Jaya) and *E. colonum* were raised in seed trays for 10 days and then transplanted into 9 cm plastic pots filled with sterile soil (loam + sand 2:1). Ten days later, when the seedlings had established, they were inoculated with 2800 freshly hatched juveniles of *M. graminicola* per pot which had been cultured on *E.*

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Table 1. Percentage decrease/increase in shoot and root growth of rice and *Echinochloa colonum* infested with *Meloidogyne graminicola* (Means of 4 Replicates).

Plants	Days	Response	Shoot		Root		Total		Lateral Axis		Total Root of Axes		No.1 No.2		Later-		Laterals		No.2		%	
			Height	Weight	Fresh Length	Length	Root Length	Root Length	Root Length	Root Length	Root Length	Root Length	Root Length	Root Length	Root Length	Root Length	Root Length	Root Length	Root Length	Root Length		Root Length
Rice	10	(-)	1.0	3.5	-	23.1	22.7	24.3	26.3	-	17.1	44.0	24.4	0	30.9	(7.5)						
		(+)	-	-	0.5	-	-	-	-	3.1	-	-	0	-	-	-	-	-	-	-	-	-
	20	(-)	3.0	2.6	-	18.7	17.5	24.5	8.2	-	28.4	-	20.3	0	-	(11.63)						
		(+)	-	-	14.9	-	-	-	-	2.8	-	58.6	-	19.5	116.7							
<i>E. colonum</i>	10	(-)	4.1	20.9	-	54.2	55.7	45.4	42.6	17.4	44.4	40.9	52.2	-	-	(27.3)						
		(+)	-	-	19.5	-	-	-	-	-	-	-	-	-	5.0	6.1						
	20	(-)	21.3	36.9	28.9	57.8	59.2	36.9	48.9	4.8	59.0	41.0	29.3	21.5	-	(12.1)						
		(+)	-	-	-	-	-	-	-	-	-	-	-	-	36.6							

*colonum* in a glasshouse. Uninoculated pots served as control. Replication was 4-fold for 2 harvests, at 10 and 20 days after inoculation (20 and 30 days after transplanting). The experiment was conducted in a completely randomised design under glasshouse conditions where the temperature ranged between 27°C and 15°C and the plants were watered every day as required.

At each harvest, shoot height and shoot and root weights were recorded; roots were washed carefully and any adhering debris picked off with forceps. The roots were analysed by a modified "line intersect" method as used by Soomro & Hague (1992a, 1992b). After root analyses, a representative root system from each treatment was selected for photography in order to illustrate the growth pattern of roots (Soomro & Hague, 1992a, 1992b).

For quantitative estimation of nematodes in roots, the roots were fixed in F.A. 4:1, stained in acid fuchsin in lactoglycerol and cleared for 24 hours in water-glycerol (50:50) solution (Bridge *et al.*, 1982). Cleared roots were then cut into 0.5-1 cm pieces, placed in a glass vial with small amount of water and macerated using a "Silverson Laboratory Homogeniser" at maximum speed for 30 seconds. The suspension was made up to a suitable volume and nematodes were counted in three 5ml aliquots under a binocular microscope (Soomro & Hague, 1992a).

## Results

**Whole plant growth:** General pattern of growth and development of rice and *E. colonum* irrespective of nematode infestation was different. Whole plant growth of both the hosts was affected by nematode invasion, but the degree of response was related to plant species (Table 1). *E. colonum* was more sensitive to *M. graminicola* invasion than rice (for example, fresh shoot weight of infested *E. colonum* 20 days after inoculation was 36.9% smaller than uninfested as compared to a 2.6% reduction in rice), yet it grew significantly bigger ( $p=0.01$ ) than even uninfested rice plants. The most significant effects of *M. graminicola* invasion were recorded 20 days after inoculation. Shoot heights, and shoot and root weights of *E. colonum* were also significantly reduced by nematode invasion (Table 1).

**Root growth and development:** The adverse effect of nematode invasion on the growth and development of rice and *E. colonum* roots was significant (Figs.1,2). Total root length, length of axes and laterals, total number of root tips and the number of primary laterals was significantly reduced by nematode invasion in both hosts. But the number of secondary laterals, axes, laterals/cm of an axis and the number of secondary laterals per one primary lateral after 20 days was increased by 58.5, 2.8, 19.5 and 116 % respectively (Table 1).

Normal branching pattern of roots in both plant species was altered and the laterals grew in clusters (Fig.3). Upto 11.6 and 27.3% laterals in rice and *E. colonum* respectively were growing on actual galls caused by the nematode invasion. When a main root was invaded and a gall formed, growth of that root was checked; this probably triggered the production of new axes, and relatively more secondary and/or tertiary laterals grew in clusters on or just above the gall. These secondary and tertiary laterals on infested roots appeared earlier than on uninfested healthy roots.

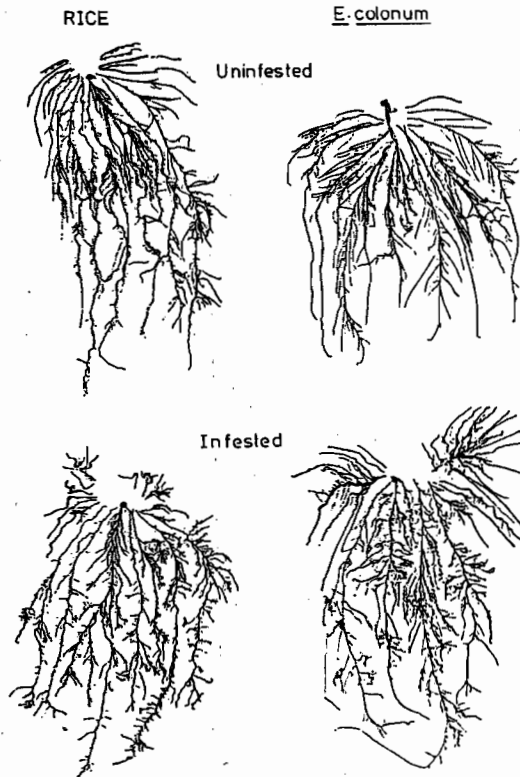


Fig.1. Root systems of rice and *Echinochloa colonum* uninfested and infested with *Meloidogyne graminicola*, 10 days after inoculation.

**Nematode invasion:** Number of nematodes invading roots was estimated only at first harvest, the results showed that total number of nematodes and the number of nematodes/1 cm of root was significantly greater ( $p=0.01$ ) in *E. colonum* than in rice (Table 2).

### Discussion

Nematodes may affect plants by altering their morphology and/or physiology. In the present study, *M. graminicola* inhibited the extension of axes, which stimulated emergence of new axes and laterals. When an axis was invaded by the nematode, it ceased to elongate, this rather triggered the emergence of new axes and lower order laterals growing in clusters on and/or around galls. These results confirm earlier reports by Soomro & Hague (1992). Results contrasting to these findings were reported by Rawsthorne & Hague (1985, 1986) who found that *Heterodera avenae* affected the seminal root growth of oats and barley by inhibiting both root extension and lateral production. The effect of *H. avenae* invasion on cereal root axes was measured by Price *et al.*, (1983) who found that the nematode hindered the extension

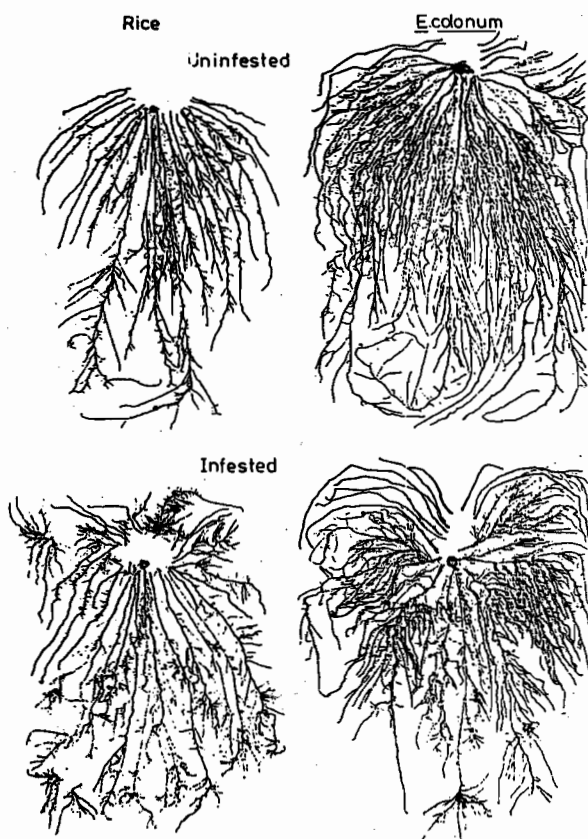


Fig.2. Root systems of rice and *Echinochloa colonum* uninfested and infested with *Meloidogyne graminicola*, 20 days after inoculation.

of axes, but after 2-3 days the roots attained the normal growth rates. Price (1979) concluded that the observed effects were a consequence of mechanical damage and/or impence of root system by nematode invasion. Similar results were reported by Soomro & Hague (1992b) on wheat infested with *M. graminicola*. However, such a result was never observed on rice or *E. colonum* infested with *M. graminicola* in the present study, which supports our previous findings (Soomro & Hague, 1992a).

Delayed emergence rather than inhibition of lateral roots has been reported by numerous workers (Slinger & Bird, 1978; O'Brien & Fisher, 1981). Some proliferation of laterals following nematode invasion, has also been observed (Bridge & Hague, 1974; Price, 1979; Rawsthorne & Hague, 1985), and the clumping of laterals was reported by Soomro & Hague (1992a), while Roy (1973) observed production of "hair like" laterals on galls caused by *M. graminicola*. In this work also, the laterals grew on galls but were relatively thick and may grow longer depending on the environmental factors (eg. temperature) and time of observation. The growth of extra laterals on galls contributed to total root numbers and their length in infested root

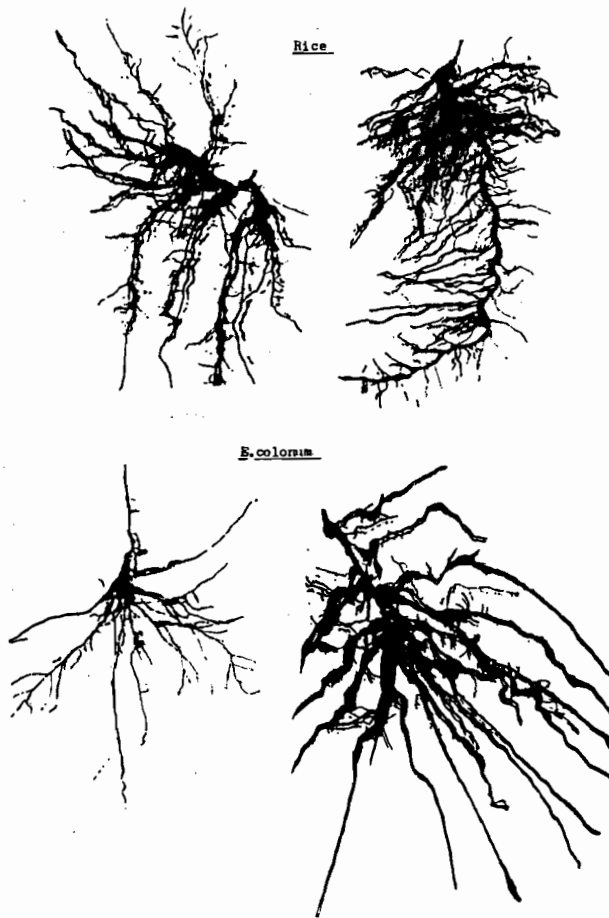


Fig.3. Rice and *Echinochloa colonum* roots showing the changes in branching pattern and clumping of laterals as a result of *Meloidogyne graminicola* invasion.

systems. Thus 20 days after inoculation there were more root tips (laterals) on infested roots (Table 1), as a result, the length of uninfested and infested roots did not differ from each other. The emergence of extra roots helps the plants grow better, and once the crucial point is overcome by the plant, it can grow and produce even though the branching pattern has been changed. Similar results were recorded by Niblack *et al.*, (1986) on soybean infested with *M. incognita* and *H. glycines*, while Shane & Barker (1986) found that the root-knot nematode *M. incognita* reduced the number and length of soybean roots.

The response of plants to parasitism by *Meloidogyne* species can be demonstrated in terms of whole plant (as in present work), but also in terms of plant cells in the immediate vicinity of parasite (Bird, 1974). Plant's response to a parasite depends on the chemical composition of the plant or tissues attacked and on the qualitative composition of nematode secretions and excretion which are related to the nematode

**Table 2. Number of nematodes in roots 10 days after inoculation (means of 3 replicates).**

	Rice	<i>Echinochloa colonum</i>
Total Number of nematodes	404.0	890.0 <sup>**</sup>
Nematodes/cm root	0.7	2.2 <sup>**</sup>
Nematodes/g root	1,816.0	1,878.0NS

\*\* = Significant at 1% level, NS = Not significant at 5% level.

species (Giebel, 1974), and, although other cogent explanations for stimulation and inhibition of growth in infested plants exist (Bridge & Hague, 1974; Rawsthorne & Hague, 1985), it is likely that growth regulatory substances play a dominant role. Thus the stimulation and clustering of laterals on roots infested with *M. graminicola* reported here could be due to the effect of some plant growth regulating substances secreted by the nematodes.

Plant growth regulators (PGRs) have been recognised to be involved in gall formation by *Meloidogyne* spp., and auxins have been identified in galls (Balasubramanian & Rangaswami, 1962; Setty & Wheeler, 1968). Viglierchio & Yu (1968) detected auxin in juveniles and egg masses of some *Meloidogyne* species, but Sandstedt & Schuster (1966) concluded that the auxins were not released from the plant tissue, nor were they secreted by the nematodes (*M. incognita*); instead, they suggested that the nematodes enabled the plant tissue to retain and use endogenous auxins, which otherwise would have been transported to the basal ends of excised tobacco tissue. PGRs may characteristically be associated with a particular nematode species, regardless of whether the healthy host normally contains auxins (Viglierchio & Yu, 1968). In other report, Viglierchio (1971) suggested that although the kind of auxins are characteristic of the species of root-knot nematode, they can be moderated by the host if it is normally high in auxin content. This may explain the differences in response of various hosts to *M. graminicola* in this work and that of Soomro & Hague (1992a).

On the other hand, auxins have been reported to stimulate lateral growth of plants (Deveson, 1987). Torrey (1986) in a review also indicated that plant hormones such as IAA, related auxins or ethylene might be involved or serve as the limiting or triggering components controlling cell division in root tissues, thus leading to initiation of lateral roots.

Ethylene has been found associated with gall formation on excised tomato roots infested with *M. javanica* (Glazer *et al.*, 1983). Stimulation of lateral growth was reported following pruning or wounding of root tissues of various plants by Biddington & Dearman (1984), and Wiedenroth & Erdman (1985). McCully (1975) suggested that when the main roots are curved in the region of lateral formation by any means, all the laterals develop on the convex side of curvature and in many cases the laterals are clumped and grow longer than single laterals. The tendency to clumping of later-

als could be due to ethylene which stimulates initiation of lateral primordia following wounding (McCully, 1975). Supporting evidence (Dawkins *et al.*, 1983) has indicated that increases in ethylene levels in impeded roots could be the result of either elevated endogenous ethylene production by the plant or by soil micro-organisms. Inhibition of axis root extension, emergence of new axes and stimulation in lateral production of barley roots occurred when the roots were exposed to ethylene, suggesting that ethylene may be involved in production of lateral roots (Crossett & Campbell, 1975).

There have been few studies of the effect of nematodes on morphology of whole root system and in future, work could be done on characterising the changes in plants caused by different types of endoparasitic nematodes in the genera of *Meloidogyne* and *Heterodera*. Further work on the effects of nematode infection on plant growth is necessary, particularly with reference to plant growth hormones such as auxins and ethylene, to find out more about root growth and the tolerance of various host plants to nematodes.

#### References

- Balasubramanian, M. and G. Rangaswami. 1962. Presence of indole compound in nematode galls. *Nature*, 194:774-775.
- Biddington, N.L. and A.S. Dearman. 1984. Shoot and root growth of lettuce seedlings following root pruning. *Annals of Botany*, 53:663-668.
- Bird, A.F. 1974. Plant response to root-knot nematodes. *Ann. Rev. Phytopath.*, 12:69-85.
- Bridge, J. and N.G.M. Hague. 1974. The feeding behaviour of *Tylenchorhynchus* and *Merlinioides* species and their effect on growth of perennial ryegrass. *Nematologica*, 20:119-130.
- Bridge, J., S.L.J. Page and S. Jordan. 1982. An improved method for staining nematodes in roots. *Rothamsted Experimental Station Annual Report for 1981*, Part I, p 171.
- Crossett, R.N. and D.J. Campbell. 1975. The effects of ethylene in the root environment upon the development of barley. *Plant and Soil*, 42:453-464.
- Dawkins, T.C.K., J.A. Roberts and J.C. Brereton. 1983. Mechanical impedance and root growth - The role of endogenous ethylene. In: *Growth Regulators in Root Development*. (Eds.) M.B. Jackson and A.D. Stead. Monograph No. 10, British Plant Growth Regulator Working Group, U.K.
- Deveson, M.R. 1987. *Effect of plant growth regulators on root growth and root/shoot integration in wheat (Triticum aestivum L.)*. Ph.D Thesis, University of Reading, Reading, England.
- Giebel, J. 1974. Biochemical mechanisms of plant resistance to nematodes: A review. *J. Nematol.*, 6:175-184.
- Glazer, I., D. Orion and A. Apelbaum. 1983. Interrelationships between ethylene production, gall formation and root-knot nematode development in tomato plants infected with *Meloidogyne javanica*. *J. Nematol.*, 15:539-544.
- Hussey, R.S. 1985. Host-parasite relationship and associated physiological changes, pp 143-154. In: *An Advance Treatise on Meloidogyne*, Vol.1, *Biology and Control*. (Eds.) J.N. Sasser and C.C. Carter North Carolina State University Graphics, Raleigh, NC USA.
- McCully, M.E. 1975. The development of lateral roots, pp 105-124. In: *The Development and Function of Roots*. (Eds.) R.G. Torrey and D.T. Clark. Academic Press, London.
- Niblack, T.L., R.S. Hussey and H.R. Boerema. 1986. Effects of *Heterodera glycines* and *Meloidogyne incognita* on early growth of soybean. *J. Nematol.*, 18:444-450.



- O'Brien, P.C. and J.M. Fisher. 1981. Ontogeny of spring wheat and barley infected with cereal cyst nematode, (*Heterodera avenae*). *Aust. J. Agric. Res.*, 32:353-364.
- Price, N.S. 1979. *Nutrient uptake and root growth in cereals as affected by cereal cyst nematode, Heterodera avenae*. Ph.D Thesis, University of Reading, Reading, England.
- Price, N.S., D.T. Clarkson and N.G.M. Hague. 1983. Root growth in oats and barley as affected by the cereal cyst nematode. *Plant Path.*, 32:377-383.
- Rawsthorne, D. and N.G.M. Hague. 1985. The relationship between size of barley roots and nematodes invasion by the cereal cyst nematode, *Heterodera avenae*. *Nematologica*, 31:79-87.
- Rawsthorne, D. and N.G.M. Hague. 1986. The effect of *Heterodera avenae* on the root system of susceptible and resistant oat seedlings. *Ann. Appl. Biol.*, 108:89-98.
- Roy, A.K. 1973. Reaction of some rice cultivars to the attack of *Meloidogyne graminicola*. *Ind. J. Nematol.*, 3:72-73.
- Sandstedt, R. and M.L. Schuster. 1966. The role of auxins in root-knot nematode-induced growth on excised tobacco stem segments. *Physiol. Plant.*, 19:960-967.
- Setty, K.G.H. and A.W. Wheeler. 1968. Growth substances in roots of tomato (*Lycopersicon esculentum* Mill.) infected with root-knot nematodes (*Meloidogyne* spp.). *Ann. Appl. Biol.*, 61:495-501.
- Shane, W.W. and K.R. Barker. 1986. Effect of temperature, plant age, soil texture and *Meloidogyne incognita* on early growth of soybean. *J. Nematol.*, 18:320-327.
- Slinger, L.A. and G.W. Bird. 1978. Ontogeny of *Daucus carota* infected with *Meloidogyne* hapla. *J. Nematol.*, 10:188-194.
- Soomro, M.H. and N.G.M. Hague. 1992a. Effect of *Meloidogyne graminicola* on root growth of graminaceous plants. *Nematol. medit.*, 20:143-147.
- Soomro, M.H. and N.G.M. Hague. 1992b. Effect of *Meloidogyne graminicola* on root growth of wheat and sorghum. *Pak. J. Nematol.*, 10:119-126.
- Torrey, J.G. 1986. Endogenous and exogenous influences on the regulation of lateral root formation, pp 31-66. In: *New Root Formation in Plants and Cuttings*. (Ed.) M.B. Jackson. Martinus Nijhoff Publishers, Dordrecht, The Netherlands.
- Viglierchio, D.R. 1971. Nematodes and other pathogens in auxinrelated plant growth disorders. *Bot. Rev.*, 37:1-321.
- Viglierchio, D.R. and P.K. Yu. 1968. Plant growth substances and plant parasitic nematodes, II: Host influence on auxin content. *Expt. Parasitol.*, 23:88-95.
- Wiedenroth, E.M. and B. Erdman. 1985. Morphological changes in wheat seedlings (*Triticum aestivum* L.) following root anaerobiosis and partial pruning of the root system. *Annals of Botany*, 56:307-316.

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