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Influence of Temperature on Encumberance of *Pasteuria penetrans* Spores and its Efficacy on Biocontrol of *Meloidogyne javanica*

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ABSTRACT

The bacterial spores of *Pasteuria penetrans* more readily encumbered on *Meloidogyne javanica* juveniles at 30° and 25.5° - 34.0°C than at 10° or 20 °C. Application of the bacterial spore powder to soil resulted in significant increase in growth characters of tomato plants. There was 37.20, 23.41, and 40.56 per cent increase in fresh and dry weight of shoot and fresh weight of root, respectively, and 78.1 per cent reduction in total nematode population at a concentration of 600 mg spore powder per kg of soil.

Key words : *Pasteuria penetrans*, spore encumberance, temperature, biocontrol *Meloidogyne javanica*

Experiments carried out to investigate the influence of temperature on spore attachment and the efficacy of *Pasteuria penetrans* (Thorne, 1940) Sayre and starr, 1985 on the biocontrol of *Meloidogyne javanica* (Treub, 1885) Chitwood, 1949 on tomato, are reported in this paper.

MATERIALS AND METHODS

The inoculum of *P. penetrans* in the form of spore powder was obtained by multiplying on *M. javanica* following the method described by Stirling and Wachtel (1980). Eight mg of spore powder was mixed with 10 ml of sterile distilled water and the slurry passed through a 100-mesh sieve to remove root fragments. The spore suspension was collected in a Petri dish (5 cm) into which 300 freshly hatched second-stage juveniles of *M. javanica* were released. A series of such Petri dishes containing spore-nematode suspension were arranged and maintained at 10°, 20°, 30° and 25.5-34.0°C (laboratory temperature). Four replications were maintained for each temperature level. After 24 hours of incubation, one ml of aliquot was taken from each Petri dish and the number of nematodes encumbered with bacterial spores was counted under a binocular microscope.

Another experiment was carried out in soil medium for a similar purpose. Fifty g of moist sterilized soil and sand mixture (1:1) was taken in a series of Petri dishes (10 cm) and spore powder was added @ 800 mg / kg soil. In addition, second-stage juveniles of *M. javanica* were also added to all the Petri dishes @ 3000 nematodes per kg soil. A small quantity of sterile water was added to soil and spore powder and nematodes were thoroughly mixed. The Petri dishes were incubated at 10°, 20°, 30° and

25.5° - 34.0°C (laboratory temperature) with four replicates under each temperature level. The soil samples were processed and active nematodes were extracted by Cobb's modified sieving and decanting method. The number of juveniles encumbered with spores was counted from an aliquot of nematode suspension.

A pot culture experiment was carried out using 15 cm diam. earthen pots containing 1.0 kg of moist sterilized soil. The spore powder was incorporated in pot soil @ 150, 300, 450 and 600 mg per kg soil. The pots were also inoculated with freshly hatched second-stage *M. j vanica* @ 3000 nematodes per kg soil and the inoculum mixed well. Four-wk-old tomato seedling cv. Pusa Ruby, were then singly transplanted in the pots. Suitable controls were included and all the treatments replicated five times. Observations on plant growth characters, nematode population in soil and roots were recorded 60 days after planting. One gram of root taken from homogenous root mixture was stained in acid fuchsin- lactophenol for 1 minute, cleared and macerated for 30 seconds in a waring blender. The suspension was made upto 100 ml and three aliquots of 2 ml were drawn. The number of eggs, juvenile and adult stages were counted. From each pot, 250 cc soil was processed by Cobb's sieving and sifting method followed by modified Baerman's funnel method. All data were subjected to analysis of variance and differences between means were evaluated for significance following modified Duncan's multiple range test (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

The rate of spore encumberance was 52.5, 70.5, 84.5, and 86.0 per cent, respectively, at 10°, 20°, 30° and 25.5°-34.0°C after 24 hours of exposure in aqueous medium (Table 1). The rate of spore encumberance in soil medium was low when compared with aqueous medium. There was only 33.8 per cent spore encumberance at room temperature and 32.5 per cent at 30°C, both being on par. As the spore encumberance on nematode body depends on the change of contact between nematode and spores, the rate of contact increases with nematode activity and density of spores. The juveniles of *M. javanica* are more active at 20-30°C than at low temperature (Wallace, 1966). Hence, the increase in the rate of spore encumberance with increase in temperature could be due to the increase in nematode motility influenced by temperature. Similarly, Stirling (1981) reported that *Bacillus penetrans* spores attached more readily to nematodes at 22.5–30°C than at 15°C.

Soil application to the bacterial spores improved plant growth characters and the growth increased with the increase in the concentration of the spore powder (Table 2). There was 37.2, 23.41 and 40.56 per cent increase in fresh and dry weight of shoot and fresh weight of root, respectively, at the highest concentration of 600 mg spore powder. Similarly, application of *P. penetrans* markedly suppressed the multiplication of *M. javanica* (Table 2). There was a progressive reduction in the rate of nematode multiplication with the increase in the concentration of spore powder. The rate of multiplication was only 4.79 at 600 mg spore powder level as against 21.9 in inoculated control. Stirling (1984) observed reduction in galling of tomato roots as well as soil nematode population by incorporating

Table 1. Effect of temperature on encumberance of second - stage juvenile of *M. javanica* by *P. penetrans* spores.

Temperature (°C)	% nematodes encumbered with spores after 24 hours of exposure[xy]	
	Water	Soil
10	52.0 a	3.7 a
20	70.5 b	17.5 b
30	84.5 c	32.5 c
25.5 - 34.0 (L. T.)	86.0 c	33.8 c

X : Mean of 4 replications
Y : Data in columns followed by a common letter are not statistically different (P = 0.5) by DMRT
L.T: Laboratory temperature

Table 2. Effect of soil application of *Pasteuria penetrans* on growth characters of tomato cv. Pusa Ruby and on multiplication of *M. Javanica*[xy]

Treatment (mg of spore powder / kg soil	Fresh weight of shoot (g)	Dry weight of shoot (g)	Fresh weight of root (g)	Total nematode population (soil+root)	Rate of multiplication (R=pf/pi)[z]
Control (Inoculated)	22.31 a	3.46 a	3.23 a	65,706 e	21.90
Control (Uninoculated	34 29 f	4.66 f	5.68 f	—	—
150	25.74 b	3.71 b	3.74 b	33,525 d	11.18
300	27.98 c	3 80 c	4.17 c	27,096 c	9.03
450	28.94 d	4.18 d	4.30 d	20,317 b	6.77
600	30.61 e	4.27 e	4.54 e	14,372 a	4.79

X: Mean of 5 replications
Y: Data in columns followed by a common letter are not statistically different (P=0.05) by DMRT.
Z: Pf = Final nematode population
Pi = Initial nematode population

the bacterial parasite in *M. javanica*-infested soil. Brown *et al.* (1985) reported that application of *P. penetrans* checked the infestation of *M. incognita* and suggested that the spores present in soil inhibited the invasion of nematodes into tomato roots. The present findings agree with the earlier reports and suggest that *P. penetrans* is effective in controlling *M. javanica* population and offer potential scope for developing it as an effective biological nematicide.

REFERENCES

BROWN, S. M., KEPNER, J. L. and SMART, Jr. G. C. 1985. Increased crop yields following application of *Bacillus penetrans* to field plots infested with *Meloidogyne incognita*. *Soil Biol. Biochem.*, **17**, 483-486.

MANKAU, R. 1975. *Bacillus penetrans* n. Comb. causing a virulent disease of plant parasitic nematodes. *J. Invertebr. Pathol.*, **26**, 333-349.

STEEL, R. G D. and TORRIE, J. D. 1980. Principles and procedures of statistics. McGraw Hill Book Co., New York 481 pp.

STIRLING, G. R. 1981. Effect of temperature on infection of *Meloidogyne javanica* by *Bacillus penetrans*. *Nematologica*, **27**, 458-462.

STIRLING, G. B. 1984. Biological control of *Meloidogyne javanica* with *Bacillus penetrans*. *Phytopathology*, **74**, 55-60.

STIRLING. G. R. and WACHTEL, M. F. 1980. Mass production of *Bacillus penetrans* for the biological control of root-knot nematode. *Nematologica*, **26**, 308-312.

WALLACE, H. R 1966. Factors influencing the infectivity of plant parasitic nematodes. *Proc. Royal, Soc. Series B (Lond.)*, **164**, 592-614.