

The role of seaweed extracts, *Ascophyllum nodosum*, in the reduction in fecundity of *Meloidogyne javanica*

Catherine A. WHAPHAM*, Teifryn JENKINS**, Gerald BLUNDEN* and Simon D. HANKINS***

* School of Pharmacy & Biomedical Sciences, Park Building

** School of Biological Sciences, King Henry Building,
University of Portsmouth, King Henry I Street,
Portsmouth, Hampshire, PO1 2DZ, U.K., and

*** Maxicrop International Ltd, Weldon Road, Corby, Northamptonshire, NN17 1US, U.K.

Accepted for publication 26 July 1993.

Summary – The effects are evaluated of commercially-available seaweed extracts, derived from *Ascophyllum nodosum*, on the fecundity of the root-knot nematode, *Meloidogyne javanica*, cultured on tomato plants (*Lycopersicon esculentum* cv. Ailsa Craig). The numbers of nematode eggs recovered from plants treated with seaweed extract were significantly reduced after one generation, compared to those plants treated with water alone. Fewer eggs were recovered from plants infected by juveniles hatched in dilutions of the seaweed extracts compared with those infected by water-hatched juveniles.

Résumé – Effets des extraits d'une algue marine sur la diminution de fécondité de *Meloidogyne javanica* – Les effets d'extraits commerciaux de l'algue marine *Ascophyllum nodosum* sur la fécondité du nématode *Meloidogyne javanica*, élevé sur tomate (*Lycopersicon esculentum* cv. Ailsa Craig) sont évalués. Après une génération, le nombre d'œufs du nématode récupérés sur les plantes traitées diminue considérablement en comparaison avec les plantes témoins, traitées avec de l'eau pure. Un plus faible nombre d'œufs sont récupérés sur les plantes infestées par des juvéniles ayant incubé dans des extraits d'algue en comparaison avec les plantes infestées par des juvéniles ayant incubé dans de l'eau pure.

Key-words : *Ascophyllum nodosum*, biological control, *Meloidogyne*, seaweed extract.

Non-pesticide control is being regarded favourably in agriculture as environmental awareness increases. Organic amendments, such as composted leaf mould (Miller, 1977) and chitin (Godoy *et al.*, 1983) have been suggested as possible alternative methods of nematode management.

Claims have been made in recent publications that the application of seaweed extracts to plants has resulted in a decreased incidence of pathogen attack. In 1977, Tarjan recorded that when *Ascophyllum nodosum* kelp extract was applied to citrus seedlings infected with *Radopholus similis* these seedlings weighed more, and had fewer nematodes, than the water control seedlings. Maxicrop and Sea-Born liquid kelp (*A. nodosum*) preparations effectively controlled sting nematodes (*Belonolaimus longicaudatus*) on centipede grass one month after application (Morgan & Tarjan, 1980). Featonby-Smith and Van Staden (1983) found that root-knot nematode infestation in tomato plants was visibly reduced in all cases when a seaweed concentrate prepared from *Ecklonia maxima* was applied as a soil drench. A similar effect was discovered by Paracer *et al.* (1987) when soil amendments of *Spatoglossum schroderi* significantly reduced root galling of tomato plants infected with *Meloidogyne incognita*, *M. arenaria* and *M. javanica*. Sea-

weed concentrate prepared from *E. maxima*, significantly suppressed the reproduction of *Pratylenchus zeae* on excised maize roots (De Waele *et al.*, 1988). More recently, Crouch and Van Staden (1993) recorded significant increases in plant growth and reduced infestation by *M. incognita* when seaweed concentrate, prepared from *E. maxima*, was applied as a soil drench.

In this study the potential role of extracts of the brown alga, *Ascophyllum nodosum*, in the control of the root-knot nematode, *M. javanica*, was investigated. Particular emphasis was given to their effects on the motility of the juvenile stages and the fecundity of the adult female.

Materials and methods

SEAWEED EXTRACTS

The brown marine alga, *A. nodosum*, provides the basis for the commercially-available seaweed extracts marketed by Maxicrop International Ltd. These aqueous alkaline extracts are formulated as Maxicrop Original, Maxicrop Triple and Maxicrop Concentrate, which contain approximately 8 %, 16 % and 24 % w/v seaweed solids, respectively. Maxicrop Triple also contains added trace elements, magnesium, iron, boron, zinc and copper being the main components.

TEST NEMATODE

Meloidogyne javanica was obtained from stock cultures maintained on tomato plants cv. Ailsa Craig. The entire root ball with soil was immersed in water and the soil removed gently, without dislodging any egg sacs. Eggs were extracted by vigorous shaking of infested roots in a 1 % sodium hypochlorite solution for three minutes. The resulting suspension was then passed through a range of different mesh sieves (Hussey & Barker, 1973). Eggs were finally collected on a fine sieve (38 μm) and washed with several changes of tap water to remove all traces of sodium hypochlorite.

HATCHING

The recovered eggs were hatched over a period of 7 days in beakers containing either water, as a control, or seaweed extract at a concentration of 3.6 % Maxicrop Original or 1.8 % Maxicrop Triple or 1.2 % Maxicrop Concentrate, which are dilutions recommended by the manufacturers for horticultural use. The eggs were aerated and left to hatch at room temperature (approximately 20 °C).

INOCULATION AND TREATMENT

A sample of 70 tomato plants, all at the four-leaf stage and planted in 450 cm³ (approximately) of John Innes No. 1 soil, was divided into seven experimental sets to give ten plants per set. Two plastic, perforated pipette tips were placed either side of the stem of each plant and pushed into the soil to a depth of approximately 2 cm. The infective dose, comprising 600 second stage juveniles (J2) of *M. javanica* per plant, was inoculated through these pipettes. Forty plants received J2 which had been hatched in tap water (water-hatched), and batches of ten plants received J2 hatched in 3.6 % Maxicrop Original, 1.8 % Maxicrop Triple or 1.2 % Maxicrop Concentrate (treatment-hatched).

Each set of plants then received 50 ml per plant of one of four treatments, applied as a root drench on a weekly basis. A sample of ten plants, infected with water-hatched J2 was treated with water, and so represented the control treatment. Twenty plants, ten infected with water-hatched J2 and ten infected with juveniles which had been hatched in 3.6 % Maxicrop Original, were treated with 3.6 % Maxicrop Original. Twenty plants, ten infected with water-hatched J2 and ten infected with juveniles which had been hatched in 1.8 % Maxicrop Triple, were treated with 1.8 % Maxicrop Triple. The final 20 plants, ten infected with water-hatched J2 and ten infected with juveniles which had been hatched in 1.2 % Maxicrop Concentrate, were treated with 1.2 % Maxicrop Concentrate. The plants were maintained in glasshouse conditions at 20 °C \pm 4 °C for 45 days post-inoculation.

The egg sacs from each plant in each experimental set were removed using 1 % sodium hypochlorite and the

total numbers of eggs recovered from the sacs were counted.

ACTIVITY ASSESSMENT

M. javanica eggs were hatched in either water or 1 % Maxicrop Original. Activity of J2 in water and 1 % Maxicrop Original was assessed using the method of Evans and Wright (1982). Briefly, 200 viable J2 were placed on the top of a sand column (height 2 cm, sand particle size 250–600 μm ; Sigma) in a polythene tube (3 \times 0.5 cm), with nylon mesh (200 μm aperture) covering the bottom end. Tubes were placed upright in a specimen tube containing 5 ml test solution (either water or 1 % Maxicrop Original). After 24 hours at 20 °C the number of juveniles that had migrated through the mesh into the outer solution was recorded.

Both methods have been subjected to repetition and consistent results have been recorded throughout the investigations. Means and standard error of the means are shown in the results section, using T-test comparisons for significance.

Results

The mean numbers of *Meloidogyne javanica* eggs recovered from each set of ten tomato plants are summarised in Figure 1. There was a significant reduction in egg recovery from plants receiving the seaweed treatments compared to the water-treated controls (Fig. 1). This difference was significant at the 5 % level for each of the three seaweed treatments. Additionally, there were significantly fewer eggs recovered from plants which had been inoculated with J2 hatched in seaweed solutions compared with plants inoculated with J2 that had been water hatched (Fig. 1). This difference was significant at the 5 % level for all three seaweed treatments. In the most extreme case, treatment of tomato plants with Maxicrop Original after infection with treatment-hatched J2, reduced the number of eggs produced after one generation 18 fold when compared to the water-treated tomato plants infected with water-hatched J2 (Fig. 1). This is a reduction of over 94 %. Maxicrop Concentrate treated tomato plants infected with water-hatched J2 showed a 1.5 fold reduction when compared to the water control, a reduction of 34.9 %.

The numbers of eggs recovered from plants which had received water-hatched J2, and which had also received the Maxicrop treatments were significantly reduced compared with those infected plants receiving water treatments alone (Fig. 1). This difference was significant at the 5 % level and had an average reduction of 42.7 %.

The effect of seaweed extracts on nematode activity was investigated using sand columns and the results demonstrate that the hatching of J2 in 1 % Maxicrop Original had a more dramatic effect on sand column migration (95 % reduction) than an environment in which the sand was soaked in 1 % Maxicrop Original (50 % reduction) (Table 1). However, both variables

produced a significant decrease in the numbers of nematodes reaching the outer solution. The nematodes hatched in 1 % Maxicrop Original solution appeared to be viable through movement seen with the use of microscope.

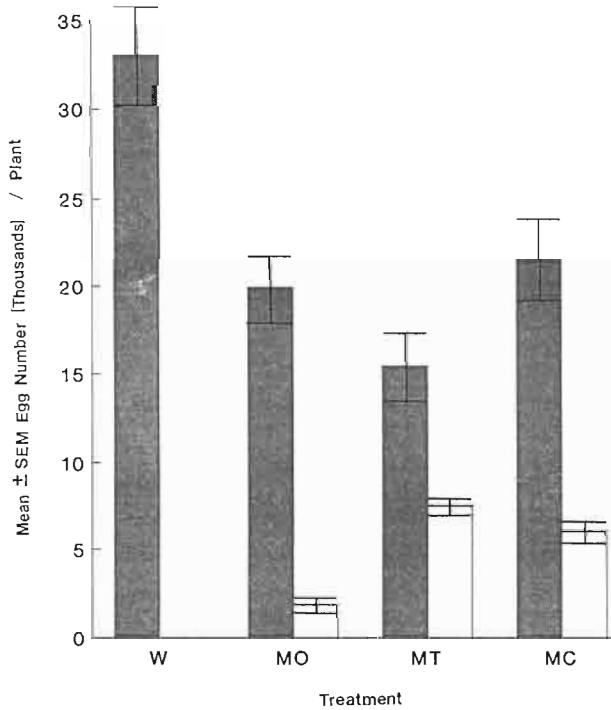


Fig. 1. The mean [± standard error (SEM)] number of eggs of *Meloidogyne javanica* per tomato plant (n = 10) after treatment for 45d with water (W), Maxicrop original (MO), Maxicrop Triple (MT) and Maxicrop Concentrate (MC). Plants had originally been inoculated with J2s which had hatched in either water (■) or the treatment solution (□).

Table 1. The mean percentage (± standard error of the mean) of J2 of *Meloidogyne javanica* reaching the outer solution after passage through sand columns (n = 5), based on an initial dose application of 200 J2.

Treatments	Water soaked sand Column	1 % Maxicrop Original soaked Column
Water-Hatched Juveniles	66.3 % ± 6.7 %	32 % ± 5 %
Treatment-Hatched Juveniles	3.0 % ± 0.9 %	1.8 % ± 1 %

Discussion

This work shows that soil drench application of seaweed extracts derived from *Ascophyllum nodosum* reduced *Meloidogyne javanica* infestation of tomato plants

compared to untreated controls. This reduction was even greater when the infective stage juveniles used as the inoculum had been hatched directly into the seaweed solutions. Plants that received water-hatched juveniles, and which were then treated weekly with seaweed extracts, also showed a significant reduction in the numbers of eggs recovered after one generation when compared with infected plants treated with water alone.

By treatment-hatching of the eggs, any effect of the treatments would be registered early on in the infective J2 stage. Inoculation with treatment-hatched juveniles produced significantly lower numbers of recovered eggs than when water-hatched juveniles were used. The activity tests support the view that reduced mobility is at least partly responsible for low levels of penetration which consequently affects egg number at the completion of the life cycle. Juveniles appeared viable when first placed on the sand columns but very low numbers migrated to the outer solutions after hatching in a seaweed environment.

Further studies are aimed at the determination of the compounds present in the seaweed extracts responsible for reducing the level of nematode populations.

References

CROUCH, I. J. & VAN STADEN, J. (1993). Effect of seaweed concentrate from *Ecklonia maxima* (Osbeck) Papenfuss on *Meloidogyne incognita* infestation on tomato *J. appl. Phycol.*, 5 : 37-43.

DE WAELE, D., McDONALD, A. H. & DE WAELE, E. (1988). Influence of seaweed concentrate on the reproduction of *Pratylenchus zaei* (Nematoda) on maize. *Nematologica*, 34 : 71-77.

EVANS, S. G. & WRIGHT, D. J. (1982). Effects of the nematocide oxamyl on the life cycle stages of *Globodera rostochiensis*. *Ann. appl. Biol.*, 100 : 511-519.

FEATONBY-SMITH, B. C. & VAN STADEN, J. (1983). The effect of seaweed concentrate on the growth of tomato plants in nematode infested soil. *Scientia Hort.*, 20 : 137-146.

GODOY, G. RODRIQUEZ-KABANA, R. SHELBY, R. A. & MORGAN-JONES, G. (1983). Chitin amendments for control of *Meloidogyne arenaria* in infested soil. II - Effects on microbial population. *Nematropica*, 13 : 63-74.

HUSSEY, R. S. & BARKER, K. R. (1973). A comparison of methods of collecting inocula for *Meloidogyne* spp., including a new technique. *Pl. Dis. Repr.*, 57 : 1025-1028.

MILLER, P. M. (1977). Reducing field populations of several plant parasitic nematodes by leaf mould composts and some other additives. *Pl. Dis. Repr.*, 61 : 328-331.

MORGAN, K. T. & TARJAN, A. C. (1980). Management of sting nematode on centipede grass with kelp extracts. *Proc. Fl. St. hort. Soc.*, 93 : 97-99.

PARACER, S., TARJAN, A. C., HODGSON, L. M. (1987). Effective use of marine algal products in the management of plant parasitic nematodes. *J. Nematol.*, 19 : 194-200.

TARJAN, A. C. (1977). Kelp derivatives for nematode infected citrus trees. *J. Nematol.*, 9 : 287.