EFFECTS OF FERTILIZERS ON PATHOGENICITY AND CHEMICAL CONTROL OF *MELOIDOGYNE INCOGNITA*

BY

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ABSTRACT

The interaction of fertilizers and carbofuran on damage caused by *M. incognita* to vegetables was studied. The damaging effects of *M. incognita* on eggplant was markedly lessened following the application of fertilizers. The growth of lettuce and carrot was enhanced but crop loss due to the nematodes was unaltered. Carbofuran significantly reduced the damage caused by *M. incognita* to these crops. The combined effect of fertilizers and carbofuran on the damaging effects of *M. incognita* to eggplant and lettuce was additive. The application of fertilizers and carbofuran alone and in combination increased the growth of eggplant infected with *M. incognita* irrespective of their time of application.

Investigations on the mechanism of action of fertilizers showed they improved the tolerance of the plants to the nematodes by enhancing the capacity of the plants to compensate for damaged roots.

A study on the mechanism of action of carbofuran showed that it inhibited the invasion of juveniles into the roots but did not have any effect on nematodes already in the roots. Hatching of *M. incognita* was delayed and the migration of second stage juveniles was affected. Behavioural responses of *M. incognita* juveniles to carbofuran on agar plates indicated that the disruption of the sensory components of the nematode was involved in its action at low concentrations.

The effect of fertilizers on damage caused by *M. incognita* on eggplant was influenced by the level and the proportion in which NPK fertilizers were applied, soil population density and plant age.

The efficacy of carbofuran against *M. incognita* was reduced by high soil temperature and by high soil organic matter.

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SECTION I

GENERAL INTRODUCTION

History and distribution of root-knot nematodes, *Meloidogyne* spp.

Root-knot nematodes, *Meloidogyne* spp., constitute a major group of plant pathogens of outstanding economic importance. The earliest record is that by Berkeley in 1855 who found galls on the roots of cucumber in an English glasshouse (Franklin, 1965). Licopoli (1875) reported the presence of root-knot nematodes within the galls on the roots of *Sempervivum tectorum* in Italy and Jobert in 1878 described galls on the roots of coffee trees in the province of Rio de Janeiro in Brazil.

Root knot nematodes were first named by Cornu (1879) as *Anguillula marioni*. During the last quarter of the 19th century and first half of the 20th century, root-knot nematodes were reported from different parts of the world and given different names: *Heterodera radicicola* (Muller, 1884); *Heterodera javanica* (Treub, 1885); *Meloidogyne exigua* (Goeldi, 1887); *Meloidogyne arenaria* (Neal, 1889); *Anguillula viates* (Lavergne, 1901) and *Caconema* spp (Cobb, 1924). Goodey, (1932) argued that root-knot nematodes should belong to the genus *Heterodera* and he named them *Heterodera marioni*. This generic name was generally accepted until the revision by Chitwood in 1949. Chitwood (1949) revived the name *Meloidogyne*..... (Goeldi, 1887) and divided it into five species and one subspecies. The early taxonomic work was reviewed by Whitehead(1968) who gave detailed descriptions of 23 species. Franklin (1972) later described 32 species. There are now 36 known species of *Meloidogyne* (Franklin, 1979).

Root-knot nematodes are world-wide in their distribution (Sasser, 1977). The most common and widespread species are *M. incognita, M. javanica, M. arenaria* and *M. hapla*.

Effects of root-knot nematode infection on root systems.

The most easily recognizable symptoms of the attack of root-knot nematodes are the galls or knots on the roots of infected plants. Heavily infected roots are shorter than those not attacked. The vascular elements are distorted and this impairs the normal translocation of nutrients and water (Muang and Jenkins, 1959; Oteifa and Elgindi, 1962; Haque *et al.*,1972). Brueske and Bergeson (1972) reported that gibberellin and cytokinin transport was reduced in roots infected with *M. incognita*.

The process of galling in plants following the infection by root-knot nematodes is brought about largely by cell hypertrophy. Gall formation starts a few hours after infection and may be induced by juveniles feeding on the root surface without actually entering the root (Loewenberg *et al.*, 1960). Galling is not essential for nematode development as has been demonstrated by Bird (1974) and Gaskin (1959). Dropkin (1972) has suggested that galling probably results from the introduction of growth regulators from the subventral glands of the second stage juveniles. Although plant growth promoting substances have been detected in *Meloidogyne* (Yu and Viglierchio, 1964) and *Ditylenchus* (Cutler and Krusberg, 1968) it is not clear whether these substances are part of the normal internal physiological processes of the nematode or whether they are exudated during feeding.

It has also been suggested that second stage juveniles of *Meloidogyne* hydrolyses plant proteins to release tryptophane which reacts with endogeneous phenolic acids to yield auxins (Setty and Wheeler, 1968), which may be responsible for cell hypertrophy.

Wallace (1973) on the other hand suggests that galling may be the result of overcompensation by the plant, in response to nematode damage. It is however not clear if these galls participate directly in reducing nutrient uptake by the plant.

Syncytia formation is another response noticeable in the roots of infected plants. This is quite a different response from gall formation. A syncytium is defined as a multinucleate mass of protoplasm formed by fusion of uninucleate cells. Syncytia are essential for the nematodes if they are to develop and mature, consequently they are considered as adaptive cellular changes (Dropkin, 1969). Enzymes particularly cellulase are believed to be responsible for the dissolution of adjoining cell walls during the formation of a syncytium. There are differences of opinion as to whether these enzymes originate from the nematodes or they are produced by the plant. Bird *et al.* (1975) believe that these enzymes are produced by the nematodes but Jones and Dropkin (1975), and Jones and Payne (1977) have suggested that the enzymes responsible for wall degradation do not originate from the nematodes but come from the plant.

There is growing evidence that syncytia function as metabolic sinks with the females functioning as the sinks (Bird and Loveys, 1975; McClure, 1977). The presence of these sinks is therefore considered a contributory factor leading to the reduction in the growth of infected plants. What is however not known is at what levels of nematode infection are syncytial response correlated with dysfunction in the infected plant.

<u>Economic importance of root-knot nematodes in vegetable production.</u>

Vegetables constitute a very sizeable portion of the diet of most humans, although the proportion in which they are consumed vary in different parts of the world. They serve mainly as sources for vegetable protein, vitamins and minerals.

The production of vegetables in virtually every part of the world is impaired to a large extent by pests and diseases including nematodes. The root-knot nematodes, *Meloidogyne* spp, are perhaps the most important of these nematodes, particularly in the tropical and subtropical areas of the world. Collectively the various species of *Meloidogyne* can attack nearly every vegetable grown. Apart from the yield losses which result from their attack, the quality of certain vegetables such as carrots and potatoes can be markedly reduced.

Estimates of losses caused by root-knot nematodes on vegetables have not been properly evaluated on a world-wide basis, but information from published papers suggests that they are very high. The problem is more serious in the tropics where the favourable climatic conditions favour rapid build-up of populations to economically damaging levels. *Meloidogyne incognita, M. javanica* and *M. arenaria* are the most important species attacking vegetables in the tropics. According to Praquin and Marchland (1970) *Meloidogyne* and other nematodes seem to be the most serious impediment to vegetable production in West Cameroons. In Senegal, Netscher (1971) has shown that yields of tomato were reduced to less than half when root-knot nematodes are present. Observations by Wilson (1962), Bridge (1972) showed that yields of tomato were reduced by 75% and 40% respectively in Nigeria, by *Meloidogyne*. Grain yields of cowpea (*Vigna sinensis* (Turner) Savi) in Nigeria were reduced by 59% (Ogunfowora, 1976). More recently Sasser (1979) and Lamberti (1979) have given estimates of losses caused by root-knot nematodes on vegetables in the tropical, subtropical and Mediterranean regions.

Estimated losses due to *Meloidogyne* spp. for selected vegetables in the tropics (Sasser, 1979).

CROP	MEAN % LOSS
Tomato	29
Egg plant	23
Okra	32
Beans	28
Pepper (all types)	15
Cabbage	26
Soyabeans (Brazil only)	26
Potato	24
Cucumber	10 - 20
Carrot	>50
Spinach	10 - 20

Field experiments done in Italy (Lamberti and Cirulli, 1970; Lamberti, 1971a, 1975) have demonstrated that severe attacks of *M. incognita* on canning tomato may cause yield losses of about 50% under climatic conditions of a Mediterranean summer. The same figure can be considered valid for table tomato production in unheated glasshouses in Malta (Lamberti *et al.*, 1976) or in Algeria (Lamberti *et al.*, 1975). Yield losses of 30 - 60% have been recorded in southern Italy (Lamberti, 1975).

Compared to the situation in the tropics and subtropics losses on vegetables due to root-knot nematodes in the temperate climate are not serious, although there are isolated cases where severe losses have been reported.

In Iceland (Siggeirsson and van Riel, 1975) have reported severe damage by *M. incognita, M. arenaria* and *M. javanica* on tomato and cucumber growing in glasshouses sometimes heated from natural sources. Damage to field grown carrots by *M. hapla* have been recorded in Sweden (Andersson, 1970); Denmark (Lindhart and Bagger, 1967); Germany (Decker, 1961); Poland (Berbec, 1972) and France (Ritter, 1972).

Apart from the direct parasitic effects of root-knot nematodes on vegetables, they are involved in disease complexes with other pathogens. They act as mechanical wounding agents, host modifiers, rhizosphere modifiers and resistance breakers. Mayol and Bergeson (1970) found that whilst the general microflora could account for only a non-significant 18% reduction in growth of tomato plants, when plants were infected with root-knot nematodes, the growth reduction was increased by 75% compared with 37% attributed to nematodes alone. Bergeson *et al.*(1970) also showed an increase in the severity of *Fusarium oxysporum* f. *lycopersici* on tomato following infection by *M. javanica.* A similar increase in severity of *Rhizoctonia solani* on okra and tomato was found following its interaction with *M. hapla* and *M. incognita* (Golden and Van Gundy, 1972). Resistance of some tomato cultivars to Fusarium wilt was broken when *M. incognita* and *M. hapla* are present,(Jenkins and Coursen, 1957).

Interaction of fertilizers with root-knot nematodes on vegetables.

The application of fertilizers is an important step in vegetable production. They are applied to supplement existing nutrients in the soil and hence ensure increased yields. Nitrogen, phosphorus and potassium fertilizers are most frequently applied as they are required in large amounts by the growing plants.

Nitrogen is essential for plant growth. It is a constituent of all proteins and nucleic acid and hence of all protoplasm. Phosphorus plays a fundamental role in the very large numbers of enzymic reactions that depend on phosphorylation. It is a constituent of the cell nucleus and is essential for cell division, development of meristem tissue and formation of reproductive structures. Potassium is important in the synthesis of ámino acids and proteins from ammonium ions. It helps in giving strength and thickness to the plant.

Although fertilizers are applied primarily to supply nutrients to the plants, information from published papers indicate that fertilizers can be used to minimize crop losses due to root-knot nematodes on vegetables. Oteifa (1952) found that lima beans infected with a relatively moderate amount of inoculum of *M. incognita* and supplied with an excessive amount of potassium made almost normal growth. The inoculated plants maintained mineral levels comparable to that of the uninoculated plants. According to Vazquez (1967) tomato plants given fertilizers at the rate of 100, 50 and 100ppm of N, P, and K respectively tolerated higher infestations and suffered less damage in terms of growth response than plants without fertilizers.

The usefulness of findings such as that of Oteifa (1952) and Vazquez (1967) are difficult to assess when fertilizers are applied at arbitrary rates. An evaluation of the influence of fertilizers on the damaging effects of root-knot nematodes on the growth of plants using application rates similar to that used under field conditions would help to assess its applicability in terms of cost.

The dual use of fertilizers both for supplying nutrients to the plants and for effectively minimizing crop losses due to root-knot nematodes would help to lower production costs, considerably. Studies are needed to determine the nature of interaction between fertilizers and root-knot nematodes on different vegetable crops in order to assess their potential for use in minimizing crop losses, when applied at economically feasible levels.

Chemical control of root-knot nematodes on vegetables.

In view of the threat posed by root-knot nematodes to the profitable production of vegetables particularly in the tropical and subtropical areas of the world, their control forms an integral part of the cultivation of these crops.

Although the most desirable and economical method of controlling these nematodes on vegetables is through the use of cultural practices and the growing of resistant varieties where feasible, the application of nematicides is still one of the most effective methods of control.

A breakthrough to large scale application of nematicides began when the nematicidal effect of the fumigant 1, 3 dichloropropene (DD) was discovered by Carter (1943). Other widely used fumigants include ethylene dibromide (EDB), first reported as a nematicide by Christie in 1945 and compounds releasing methylisothiocyanate (MIT). DBCP, 1, 2 dibromo-3-chloropropane reported by McBeth in 1954 as nematicidal in the Nemagon formulation was perhaps the most widely used fumigant in the tropics until it was recently banned. DBCP is relatively non phytotoxic to most vegetable crops and in Nigeria this was the most widely used nematicide for the control of root-knot nematodes on vegetables.

Many of the newly developed non-fumigant organophosphate and carbamate nematicides are systemic and are available in granular formulations and are relatively nonphytotoxic. They could therefore replace DBCP for the control of root-knot nematodes on vegetables in Nigeria, provided they are effective, safe to use, and cheap.

Organophosphate and carbamate nematicides have been used successfully for the control of root-knot nematodes on vegetables. Averre *et al.* (1974) found that aldicarb (Temik), carbofuran (Furadan) and ethroprop (Mocap), methomyl (Lannate), oxamyl (Vydate), fensulphothion (Terracur) and phenamiphos (Nemacur) were all active against *M. incognita* when incorporated into the soil before transplanting sweet potato. Reddy and Rao (1975). reported that aldicarb, oxamyl, carbofuran and fensulphothion controlled *M. incognita* and increased yields of soyabeans. The efficacy of oxamyl, carbofuran, aldicarb, fensulphothion, phenamiphos and ethroprop for the control of *M. incognita* on cucumber has also been reported (Rhoades, 1975). One of the factors which limit the extensive use of organophosphate and carbamate nematicides is their high mammalian toxicity. Apart from the residues which might be left in the crop, the greatest occupational hazard from pesticides is from dermal exposure. Only through gross carelessness does exposure from oral ingestion occur. Of the non volatile nematicides, carbofuran is one of those that possess a very low acute dermal toxicity (LD_{50} > 10,200mg/kg body weight for rabbits), which reduces the risk of poisoning through dermal exposure. Carbofuran is a carbamate pesticide having a broad spectrum utility as a nematicide, insecticide and acaricide. The use of this pesticide for the control of root-knot nematodes attacking vegetables could also help to reduce production costs by eliminating or minimizing the further application of insecticides.

The nematicidal efficacy of carbofuran against root-knot was first reported by Di Sanzo (1969). Although this nematicide has been used successfully to increase yields of vegetables in fields infested with root-knot nematodes (Averre *et al.*,1974; Sivakumar *et al.*,1974, 1976; Kinlock 1974; Overman and Jones, 1975) the actual extent to which it reduces nematode damage is uncertain. The increased yields obtained in field experiments might result from the control of not only the nematodes but soil and aerial pests. Its actual potential for reducing crop losses due to *Meloidogyne* can be better assessed from experiments carried out with plants grown in inoculated sterilized soil and with a suitable control also treated with the nematicide but not inoculated for comparison.

The relationship between carbofuran and fertilizers when both are applied in vegetable production has not been studied. Fertilizers may enhance or reduce the nematicidal efficacy of carbofuran. Yoshida *et al.*(1974) found that the control of *Aphelenchoides besseyi* on rice by diazinon was less when it was applied simultaneously with fertilizers to seedbeds. Yein *et al.* (1977) found that the control of *M. incognita* on mung, *Phaseolus aureus* Roxb. with aldicarb was significantly greater when applied with phosphorus and nitrogen fertilizers than when aldicarb was applied alone.

Knowledge of the possible interaction between fertilizers and carbofuran will help to explain the resultant effects obtained when both are applied in vegetable production.

More information is also needed on the interaction of carbofuran with root-knot nematodes on different vegetables,on the mechanism by which it controls *Meloidogyne* and factors affecting its efficacy.

Chemical and physical properties of carbofuran.

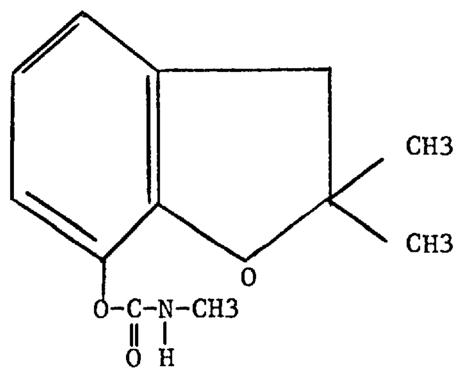
Chemical name : 2,3-Dihydro-2,2-dimethyl-7-benzo furanyl methylcarbamate.

Empirical formula: $C_{12}H_{15}NO_3$

Registered trademark: Furadan, Curaterr.

Common name: carbofuran

Chemical structure:



Appearance: White crystalline solid when pure

Odour : None when pure

Specific gravity : 1.180 at 20/20°C

Melting point : pure - 153-154°C

Technical - 150-152°C

Vapour pressure: Very low 1.1 x 10^{-4} at 50°C

Flammability : Non flammable

Stability : stable in acid media, unstable in alkaline media.

Solubility : 250 - 700ppm at 25°C in water and less than

5% in most common organic solvents.

12% in methylene chloride

15% in acetone.

Toxicology: Acute dermal toxicity (rabbits)

	LD_{50} (mg/kg)
Carbofuran (technical material)	> 10,200
Furadan 75 Dustless Base	3,400
Furadan 10G	> 10,200

Object of this study.

Root-knot nematodes are the most destructive nematode pests of vegetables in my country, Nigeria (Bridge, 1972; Bos, 1977; Ogunfowora, 1976). The most desirable and economical method of controlling these nematodes in a developing country like Nigeria, where farmers lack sufficient capital for investment on inputs, will be through the use of cultural practices and the growing of resistant varieties, where feasible. In situations where nematicide application is required the use of pesticides with broad spectrum utility as a nematicide and insecticide would be highly desirable.

The use of fertilizers and carbofuran look promising for inclusion in designing such a management programme for the control of root-knot nematodes on vegetables, where the emphasis is on economics.

Against this background, studies were carried out on the effects of fertilizers on pathogenicity and chemical control of *M. incognita* with carbofuran in relation to the damage caused by these nematodes to different vegetables.

SECTION 2

GENERAL MATERIALS AND METHODS

NEMATODES - SOURCE AND ESTABLISHMENT OF PURE POPULATION

The population of *Meloidogyne incognita* (Kofoid and White, 1919) Chitwood, 1949, used for this work was originally collected from vegetables in Malawi. A pure culture of this species was established from a population of *Meloidogyne* spp. maintained on carrot in a tropical glasshouse of Imperial College Field Station.

To establish a pure culture of *M. incognita* egg masses were collected from the roots of infected carrot plants. Each egg mass was placed in a separate watchglass. Three week old seedlings of tomato, variety Moneymaker, were transplanted singly into 22.5cm plastic pots filled with heat sterilised soil. Each seedling was inoculated by placing one egg mass below the roots in the planting hole at transplanting. Six weeks after inoculation, mature females were extracted from the roots of infected plants and perineal patterns prepared. Identification to species was based on the nature of the perineal pattern.

Egg masses were obtained from the roots of plants having females with perineal patterns typical of *M. incognita*. These egg masses were subcultured into several pots of tomato to provide ready sources of nematodes for work whenever required.

NEMATODE INOCULUM AND INOCULATION TECHNIQUE

Egg masses were picked from washed infested roots of tomato plants and placed on small 90µ nylon mesh sieves in a watch glass, containing distilled water. The level of distilled water was adjusted until the bottom surface of the sieve was immersed. The watch glass was covered with a glass plate and kept in a constant temperature room maintained at $33^o \pm 2^oC$. Hatched juveniles which were collected within the first seven days were the only ones used.

A suspension of these freshly hatched juveniles was poured into a beaker and made up to $100cm^3$ with water. Three $1cm^3$ aliquots of this larval suspension were counted to determine the average number of juveniles per cm^3. On the basis of this count, aliquots containing the known number required for inoculation was added with the help of a syringe into four small holes about 3cm deep made around the base of the plant.

ESTIMATING NEMATODE POPULATION IN ROOTS

The root systems of infected plants were carefully removed from the pots and washed in running tap water to remove adhering soil. Excess water was removed by pressing lightly between two blotting papers before roots were weighed.

The roots were cut into small pieces about 5mm long. One gram representative samples taken randomly from the cut root pieces were stained in boiling 0.1% cotton-blue in lactophenol.

After cleaning in cold, clear lactophenol for at least 48 hours excess stain was washed off under a stream of water. The stained roots were macerated for 30 seconds at half speed in a Waring blender containing $50cm^3$ of water. This process released the nematodes from the roots.

The suspension of nematodes was poured into a beaker and made up to $100cm^3$ with water. The number of nematodes in $10cm^3$ aliquots withdrawn from this suspension after thorough mixing, were counted with the aid of a dissecting microscope. On the basis of these counts, estimates of the number of nematodes per gram of root were obtained. From this count it was possible to obtain estimates of the root population of nematodes per root system. In some cases where the root systems were small, whole root systems were macerated.

ESTIMATES OF THE POPULATION OF JUVENILES IN THE SOIL

Second stage juveniles in soil were extracted using a modification of the method described by Whitehead and Hemming (1965). All the soil from each pot was poured into a tray and mixed thoroughly. After all root pieces had been removed two 200g subsamples were taken from the bulk sample and each one was spread as a thin layer on a single sheet of Kleenex tissue. Extraction was continued for 48 hours, allowing active juveniles to pass through the tissue into the water in the tray. The suspension was collected in a beaker and allowed to stand for 24 hours. The supernatant was poured off. The number of nematodes were estimated from $10cm^3$ aliquots withdrawn from the suspension, after thorough mixing.

APPLICATION OF FERTILIZERS

The calculated amount of fertilizers required per pot was mixed with the soil with a hand trowel at transplanting or sowing. As fertilizers are usually applied as side dressing in eggplant production in Nigeria, the amount added per pot was based on the plant population per hectare. For example, eggplant is planted at the rate of 41,000 plants per hectare in the same way as tomato. Thus to apply 50kg of nitrogen per hectare will mean giving each plant $(\frac{50 \times 1000}{41,000} g) = 1.2g$.

For post plant application of fertilizers, the calculated amounts were added to four small holes about 4cm deep made around the base of the plant. The addition of 18.29g of NPK (777) fertilizers at transplanting, followed by half this amount at 6 weeks is referred to in this thesis as the 'normal' rate of application.

This term also applies to the addition of only 18.29g of the fertilizers to the pot at transplanting if the experiment was terminated before six weeks.

The 'no fertilizer' treatment referred to in the text actually represents a low level of N, P and K as all pots were given an initial dose of 1.5g/kg of soil at the start of each experiment, except where otherwise stated.

POTTING OF SOIL

A mixture of heat sterilized sand, loam and peat in the ratio of 3 : 2 : 1 was the type of soil used for all the experiments except where otherwise stated.

Prior to potting, each pot was washed thoroughly and lined at the bottom with a thin polythene sheet, leaving only two small holes at the bottom. Each pot was then placed into each plastic saucer.

These precautions were taken to prevent excessive loss of fertilizers and pesticide during watering, which was done twice daily in the summer months.

NEMATICIDE APPLICATION

Carbofuran was applied as Furadan 10G supplied by FMC. This is a granular formulation of carbofuran containing 10% active ingredient. The amount of carbofuran required per given weight of soil was mixed with the soil in a polythene bag before being poured into the plastic pots. For post plant application, soil around the roots was loosened and the nematicide mixed with the soil and watered-in immediately.

The amount of nematicide to be added to a known weight of soil was based on the weight of a hectare-furrow slice to a depth of 20cm which is about 2 million kg for a typical soil (Thompson and Troeh, 1978). Thus if carbofuran is to be added to 5kg weight of soil at the rate of 8kg active ingredient per hectare which equals 80kg Furadan 10G, the amount required will be calculated as follows:

5kg of soil will require $\frac{80 \quad X5}{2,000,000}$ = 200mg of Furadan 10G.

DETERMINATION OF SOIL pH

The pH of soil was determined using the method described by Piper (1950). Twenty grams of soil was placed in a conical flask and 100cm^3 of deionized water was added. The flask was shaken mechanically for one hour before the pH was measured using a pH meter. In experiments where fertilizers and nematicides were added to soil, additional pots were included for pH determination two days after the start of the experiment.

ROOT-KNOT RATING

The degree of root galling was assessed using either of the methods stated below.

(i) A rating system modified from the method described by Daulton and Nusbaum (1961).

0 - free from galls

1 - trace of galls; less than 5 galls per root system

2 - very slight galling; 5-25 galls per root system

3 - moderate galling; 26-100 galls per root system

4 - galls numerous, but mostly discrete

5 - galls numerous, mostly coalesced

6 - very heavy galling, large galls predominate.

(ii) A rating system described by Bridge and Page (1977).

0 - no galls

1 - few small knots, difficult to find

2 - small knots only but clearly visible. Main roots clean

3 - some larger knots visible. Main roots clean

4 - larger knots predominate, but main roots clean

5 - 50% of roots infested. Knotting on parts of the main roots. Root system reduced.

6 - knotting on main roots

7 - majority of main roots knotted

8 - all main roots knotted. Few clean roots visible

9 - all roots severely knotted. Plant usually dying

10 - all roots severely knotted. No root system. Plants usually dead.

STATISTICAL ANALYSIS

Treatment means were compared using the analysis of variance technique. The two-way analysis of variance was used to test for interactions between various factors.

SECTION 3

3.0 EFFECTS OF FERTILIZERS AND CARBOFURAN ON GROWTH OF VEGETABLES INFECTED WITH *M. INCOGNITA*

INTRODUCTION

Eggplant, *Solanum melongena* L., Lettuce *Lactuca sativa* and carrots, *Daucus carota* represent the three main types of vegetables. Eggplant is a fruit vegetable while lettuce and carrots are leaf and root vegetables respectively.

Root-knot nematodes, *Meloidogyne incognita* pose a serious threat to their profitable production in different parts of the world. Their economic importance as severe pests of eggplant has been reported by several workers. *M. incognita* was observed by Bridge (1972) as a serious pest of eggplant in irrigated fields in northern Nigeria. In southern Italy, it is frequently attacked by *M. incognita* resulting in yield losses of 30-60% (Lamberti, 1975). *M. incognita* is an important pest of eggplant in the Sudan (Yassin, 1974). Bhatti and Ramesh (1977) reported yield losses of 27.3% on eggplant in India, due to *M. incognita*. These nematodes can also severely reduce the production of lettuce and carrots in the tropics and subtropics (Lamberti, 1979). They are particularly destructive to carrot as they not only reduce the production of tap roots but also reduce their marketable value by causing abnormal root growth (Lamberti, 1971b).

Fertilizers containing nitrogen, phosphorus and potassium are often applied during the cultivation of these vegetables to ensure good yields. Studies have not been done to determine the relationship between NPK fertilizers, applied at rates similar to that used for vegetable production, and the damaging effects of *M. incognita* to the crops.

The application of the newer granular nematicides e.g. carbofuran could provide an economical way of reducing crop losses due to *M. incognita* and insects on vegetables.

The studies reported in this section were designed:

(i) to evaluate the effects of fertilizers and carbofuran on the damaging effects of *M. incognita* on eggplant, lettuce and carrots.

(ii) to study the relationship between fertilizers and carbofuran when applied together to ensure the good growth of vegetables infected with *M. incognita*.

The influence of time of application on their effects when applied alone and in combination to eggplant infected with *M. incognita*, was also studied.

3.1 INFLUENCE OF TIME OF APPLICATION ON THE EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF EGGPLANT

In studies concerning the effects of fertilizers and carbofuran on the growth of vegetables infected with root-knot nematodes, their effectiveness as post-plant treatments should be considered. Fertilizer practices, in terms of time of application, vary in different parts of the world. For example, fertilizers are at times applied as post-plant side dressings in vegetable production in the tropics (Purseglove, 1968). Often root-knot is not detected in a field before a susceptible crop is planted and in this situation nonphytotoxic nematicides can be most effectively used as post-plant treatments. Carbofuran has been used successfully to increase the yields of tomato in soil infested with root knot nematodes when applied as post-plant treatments (Sivakumar *et al.*, 1974, 1976).

The individual effects of fertilizers and carbofuran when applied together should therefore be assessed at the two possible times of application.

The following experiment was set up to determine the influence of time of application on the effects of fertilizers and carbofuran when applied alone and in combination on the growth of eggplant infected with *M. incognita*.

MATERIALS AND METHODS

Forty-eight, 22.5cm, plastic pots were each filled with 5kg of heat sterilized soil. A compound fertilizer 'Growmore' containing 7% N, 3%P and 5.8%K (N7, $P_2O_5$7; K_2O7) was used. This was added to the pots at a rate corresponding to 750kg per hectare, to give 52.5kg of N, 22.5 of P and 43.5kg of K (see General Materials and Methods for calculations). At this rate of application 18.29g was added to each pot prior to transplanting. At six weeks after transplanting, half of this amount was also added to the pots given fertilizers at transplanting. The rate of fertilizer application was similar to that used on eggplant and other vegetables in Nigeria.

Carbofuran was applied as Furadan 10G at the rate of 8kg a.i./ha within the range recommended by the manufacturers for vegetable production.

Treatments were as follows:-

(i) Fertilizers only at transplanting

(ii) Fertilizers applied 2 weeks after transplanting

(iii) Nematicide only at transplanting

(iv) Nematicide only 2 weeks after transplanting

(v) Fertilizers and nematicide applied together at transplanting

(vi) Fertilizers applied at transplanting and nematicide at two weeks later

(vii) Nematicide applied at transplanting and fertilizers at two weeks later

(viii) None (control)

Four week old seedlings of eggplant (local Nigerian variety) were transplanted singly into each pot and inoculated with 2,000 freshly hatched second stage juveniles. A preliminary experiment had shown that this number of second stage juveniles was capable of significantly reducing the growth of this variety of eggplant of similar age.

Each treatment was replicated four times. Two additional replicates per treatment were included for observations at six weeks after inoculation. Pots were arranged in a completely randomized design on a glasshouse bench where temperatures ranged from 16^{o}-$41^{o}C$. Pots were watered when necessary. Soil pH was determined for each treatment (see General Materials and Methods).

Six weeks after inoculation, the two additional replicates per treatment were harvested. The fresh weight of shoots and roots were taken. The number of galls and females per root system were estimated. The number of eggs per egg mass was also assessed.

The remaining plants (4 replicates) were harvested 90 days after inoculation, when fresh weight of tops (stem, leaves and fruits) were taken. Weights of matured fruits per treatment alone were also taken. The fresh weight of roots was taken and the roots assessed for galling according to the method described by Bridge and Page (1977). The soil populations of the second stage juveniles were estimated.

RESULTS

Initial results from plants 42 days after inoculation

The results given in Table 3.2.1 were based on two replicates and were therefore not analysed.

The application of fertilizers and carbofuran alone or in combination increased top weight of plants. This was irrespective of their time of application. The root weights of plants were also increased by the application of fertilizers alone or when applied with carbofuran.

Table 3.2.1 Effects of times of application of fertilizer and carbofuran on eggplant growth and *M. incognita* populations 1. Initial results from plants 42 days after inoculation

Treatment	Fresh weight of tops in g [1)]	Fresh weight of roots in g	No. of galls per root system	No. of galls per gram of root	No. of Females per root system	No. of eggs per egg mass
1 Fertilizers only at transplanting	69.2	23.7	2561	108.0	1156	317.5
2 Fertilizers only 2 weeks after transplanting	56.7	18.3	2489	136.0	1244	325.0
3 Nematicide only at trans-transplanting	66.8	12.6	292	23.20	159	300.0
4 Nematicide only 2 weeks after transplanting	42.9	11.1	1440	129.7	732	380.0
5 Fertilizers + Nematicide applied together at transplanting	75.3	14.6	986	67.5	221	342.5
6 Fertilizers at transplanting + Nematicide 2 weeks later	71.2	18.6	1646	88.5	804	332.5
7 Nematicide at transplanting + Fertilizers 2 weeks later	79.2	19.5	843	43.2	194	377.5
8 No fertilizer No nematicide applied (control)	26.3	14.8	2269	153.3	1302	352.5

1) Mean of two replicates

The number of galls per root system was increased by fertilizer application but the number of galls per gram of root was less when compared with those on control plants (Table 3.2.1). The number of galls and females per root system was reduced when carbofuran was applied alone or with fertilizers. There were fewer galls and females per root system when carbofuran was applied alone.

The fecundity of the females based on the number of eggs per egg mass was not adversely affected by any of the treatments when compared with controls (Table 3.2.1).

<u>Data taken at harvesting 90 days after inoculation</u>

Based on fresh weight of tops, the application of fertilizers and carbofuran alone and when applied together significantly ($P < 0.01$) increased the growth of plants (Table 3.2.2). The greatest top weights were obtained when fertilizers were applied with carbofuran and this was irrespective of time of application of fertilizers and carbofuran (Table 3.2.2).

There was no difference in the growth of plants in soils given fertilizers and treated with carbofuran when the time of fertilizer application was varied ($P > 0.05$). But significantly ($P < 0.01$) greater top weights were obtained when the nematicide was applied at transplanting to fertilized soil than at two weeks later.

Only plants grown in soil to which fertilizers were applied alone or in combination with nematicide had mature marketable fruits at harvesting (Table 3.2.2). Despite good control of nematodes (Table 3.2.1) plants in soil treated with carbofuran alone, did not produce marketable fruits apparently due to the poor nutrient status of the soil.

Plate 1: Influence of time of application of fertilizers and carbofuran on the growth of eggplant infected with *M.incognita*

(35 days after inoculation).

A - No fertilizers no nematicide applied

B - Nematicide alone applied two weeks after inoculation

C - Nematicide only applied at transplanting

D - Fertilizers only applied two weeks after inoculation

E - Fertilizers only applied at transplanting

F - Fertilizers and nematicide applied together at transplanting

G - Fertilizers applied at transplanting, nematicide two weeks later

H - Nematicide applied at transplanting, fertilizers two weeks later


a
b
c
d
e
f
g
h

Table 3.2.2 Effects of times of application of fertilizer and carbofuran on eggplant growth and *M. incognita* populations 2. At harvesting

Treatments	Fresh weight of tops (including fruits) in g	Weight of mature marketable fruits	Fresh weight of roots	Gall ratings	Final soil population of juveniles per 200g of soil
1 Fertilizers only at transplanting	142.7[1]	83.5	50.93	7.25	2394
2 Fertilizers only 2 weeks after transplanting	125.2	65.0	49.05	7.50	2060
3 Nematicide only at transplanting	87.2	0	38.70	7.25	1070
4 Nematicide only 2 weeks after transplanting	69.6	0	28.55	7.50	1114
5 Fertilizer + Nematicide applied together at transplanting	223.7	139.9	52.10	7.00	2051
6 Fertilizer at transplanting + Nematicide applied 2 weeks later	181.9	108.3	49.90	7.25	2479
7 Nematicide at transplanting + Fertilizer 2 weeks later	239.3	150.0	50.50	7.00	2693
8 No fertilizer no Nematicide applied (control)	37.2	0	22.43	8.25	1024
L.S.D. at 5%	23.1	24.4	5.7	0.6	801.5
L.S.D. at 1%	31.3	33.8	7.8	0.8	1086.2

1 Mean of four replicates

The marketable yields of plants grown in soil given fertilizers and treated with nematicide were significantly ($P < 0.01$) higher than the yields obtained for plants given only fertilizers. The marketable yields of plants in fertilized soil and treated with nematicide at transplanting were significantly ($P < 0.05$) higher than the yields obtained for plants in fertilized soil but treated with nematicide at the later date (Table 3.2.2).

The top weights of plants given fertilizers alone at the two times of application were significantly ($P < 0.01$) greater than those of plants given carbofuran alone.

There was no significant ($P \ngtr 0.05$) difference between the top weights of plants given fertilizers alone at the two times of application. Similarly, no difference was found between the top weight of plants in soil treated with nematicide alone at the two dates of application ($P > 0.05$).

The application of fertilizers and carbofuran alone or in combination significantly ($P < 0.05$) increased root weight of plants over those of control (Table 3.2.2). The greatest root weights were obtained for plants grown in soil given fertilizers alone or in combination with carbofuran. The root weights obtained from these treatments were greater than those of plants in soil treated with carbofuran alone ($P < 0.01$).

The roots of all plants inoculated with nematodes, irrespective of treatment and time of application, were severely galled at harvesting (Table 3.2.2). The roots of plants in soil without fertilizers and carbofuran were more severely galled when compared with the ratings obtained for plants given the other treatments ($P < 0.05$). There was no difference between the gall ratings obtained for plants given the other treatments ($P > 0.05$).

Based on the number of second stage juveniles, the final soil population for the various treatments were all higher than the initial population (Table 3.2.2). The highest final soil populations were found in pots to which fertilizers were added alone or in combination with nematicide. The final population of juveniles in soil treated with carbofuran and given fertilizers was significantly ($P < 0.05$) higher thanthat found in soil treated with carbofuran only.

Time of application of fertilizers and carbofuran had no significant influence on the final soil populations in soil given the combined application of fertilizers and carbofuran. There was no difference between the final soil populations in soil treated with carbofuran alone and in soil without fertilizers and not treated with carbofuran ($P > 0.05$).

3.2 EFFECTS OF FERTILIZERS AND CARBOFURAN APPLIED SINGLY AND IN COMBINATION ON THE GROWTH OF EGGPLANT INFECTED WITH *M. INCOGNITA*

Although the results obtained from the previous experiment (Section 3.1 Table 3.2.2) showed significant increases in the growth of infected plants, it was not possible to determine if, and to what extent, the damaging effects of the nematodes on the crop was affected.

The inclusion of plants given similar treatments but not inoculated, for comparison, could throw some light on the relationship between the application of fertilizers and carbofuran when applied singly and in combination and the damage caused by *M. incognita* on eggplant. An experiment designed in this way would also help to show if carbofuran at the rate applied stimulated plant growth. The possible stimulatory effect of carbofuran on plant growth was reported by Reddy (1975).

The following experiment was designed to evaluate the effects of fertilizers and carbofuran when applied singly and in combination on the growth of eggplant infected with *M. incognita*.

MATERIALS AND METHODS

Four week old seedlings of eggplant were transplanted singly into 22.5cm plastic pots filled with 5kg of heat-sterilized soil. Prior to transplanting, fertilizers (18.29g/pot) and carbofuran (8kg a.i./ha) were added singly and in combination to the pots. Details of the various treatments are given in Table 3.3.1.

One half of these pots were inoculated at the rate of 2000 freshly hatched second stage juveniles of *M. incognita* per pot. An equal number of uninoculated pots served as control.

Pots were arranged in a completely randomized design on a glasshouse bench where the temperature ranged from 18°-36°C. At six weeks after inoculation, fertilizers were added to those pots which had been given fertilizers initially at the rate of 9.18g/pot.

Sixty-five days after inoculation, the experiment was terminated. The fresh weight of tops and roots were taken and the degree of root galling assessed using a modification of the method described by Daulton and Nausbaum (1961). The data obtained on fresh weight of tops and roots were analysed using the analysis of variance technique to compare the effects of different treatments. The two-way analysis of variance was used to test for interactions between the various factors- fertilizers, carbofuran and nematodes.

RESULTS

The application of fertilizers and carbofuran alone and in combination significantly ($P < 0.01$) increased the top weights of infected plants (Table 3.3.1). Compared with uninfected plants given the same treatment, the top weight of infected plants in soil without fertilizers was reduced by 69.8%, but in soil given fertilizers a reduction of only 23.3% was obtained. Similarly the reduction in top growth of plants in soil treated with carbofuran alone and in combination with fertilizers were 33.8% and 11.8% respectively. (Table 3.3.1) The reduction in top growth when carbofuran and fertilizers were applied together was not significant ($P > 0.05$). The interaction, fertilizers x nematodes was significant ($P < 0.05$) for top weight

Table 3.3.1 Effects of fertilizers and carbofuran applied singly and in combination on the growth of eggplant infected with *M. incognita*

Treatments	Fresh weight of tops in g 1)	% Decrease in in top weight with nematodes	Fresh weight of roots in g	% Decrease or increase in root wt. with nematodes	Galling Index
1 a) Fertilizers + Nematodes	74.2	23.3	41.4	+25.8	5.2
b) Fertilizers - Nematodes	96.7		32.9		0
2 a) Carbofuran + Nematodes	42.8	33.8	21.3	+7.0	4.0
b) Carbofuran - Nematodes	64.7		19.9		0
3 a) Fertilizers + Carbofuran + Nematodes	92.8	11.5	38.2	+18.6	4.0
b) Fertilizers + Carbofuran - Nematodes	104.8		32.2		0
4 a) No fertilizer No nematicide + Nematodes	18.9	69.8	12.2	-31.1	5.4
b) No fertilizer No nematicide - Nematodes	62.6		17.7		0
L.S.D. at 5%	14.1		3.7		1.1
L.S.D. at 1%	19.1		5.1		1.6

1) Mean of four replicates

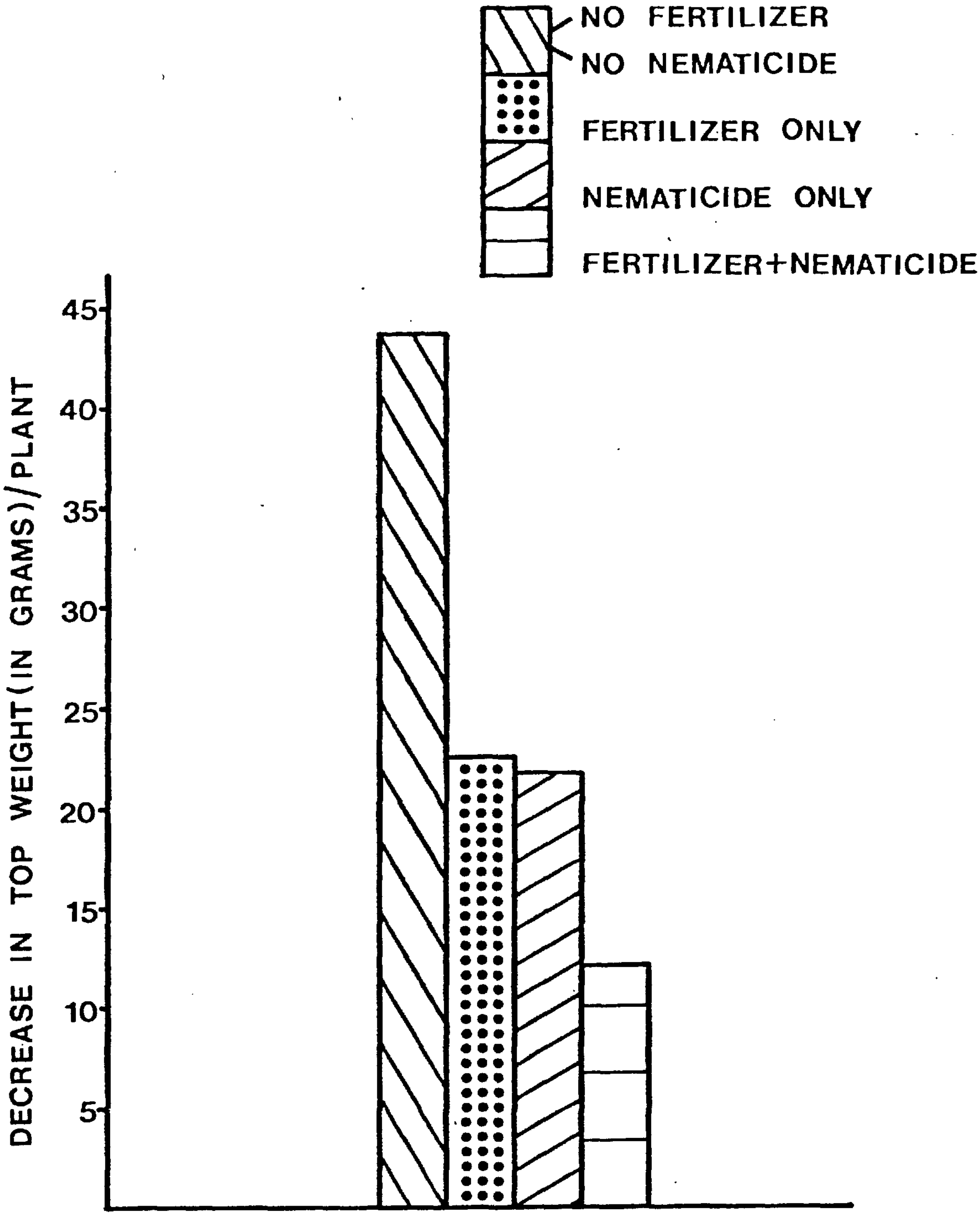


Fig. 3.2.1. Reduction in top growth of eggplant infected with *M. incognita*.

(Differences between the mean top weights of uninfected and infected plants).

The interaction, carbofuran x nematode was also significant ($P < 0.01$) but fertilizer x nematicide interaction was not significant ($P > 0.05$).

The incidence of root galling was significantly ($P < 0.05$) reduced by the application of carbofuran alone or in combination with fertilizers (Table 3.3.1).

There was no difference in the incidence of galling on the roots of plants in soil given fertilizers and in unfertilized soil ($P > 0.05$).

The root weights of infected plants in soil given fertilizers and carbofuran alone or in combination were greater than those of uninfected plants but the root weight of infected plants in unfertilized soil was significantly ($P < 0.05$) reduced (Table 3.3.1.).

The application of carbofuran did not stimulate the growth of the plants.

3.3 EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF LETTUCE AND CARROT INFECTED WITH *M. INCOGNITA*

Both lettuce and carrot are usually seeded directly into the field in tropical countries. As the application of NPK fertilizers minimized considerably the damaging effects of *M. incognita* on eggplant (Section 3.2), it was considered of importance to also determine the effects of their application on the growth of lettuce and carrot infected with *M. incognita*, because of the different cultivation practices. In the tropics, the use of carbofuran could provide an economical way of protecting the young seedlings from damage by root-knot nematodes during the early stages of growth when they are most vulnerable.

The following experiment therefore, sets out to evaluate the effects of fertilizers and carbofuran on the growth of lettuce and carrot infected with *M. incognita*.

MATERIALS AND METHODS

Seeds of lettuce, variety 'Avondefiance' and carrot variety 'Nantes champion scarlet horn' were sown in 22.5cm plastic pots containing 5kg of heat-sterilized soil.

One week after germination, the seedlings of lettuce and carrot were thinned to one and three per pot respectively, and inoculated at the rate of 1,200 freshly hatched juveniles per pot. An equal number of pots not inoculated served as controls. Prior to planting, NPK fertilizers were added to some of the pots at the rate of 18.29g per pot. Half of this amount was added to these pots six weeks after inoculation. Carbofuran was applied at the rate of 8kg a.i./ha

(200mg/pot). Details of the various treatments are given in Tables 3.4.1 and 3.4.2. Pots were arranged in a fully randomized design in the glasshouse where the temperature ranged from 13^{o}-39^{o}C.

Carrot plants were thinned to one per pot, five weeks after sowing to reduce plant population as their growth was observed to be slow.

Plantswere harvested sixty-five days after planting. The fresh weight of leaves and roots of lettuce plants were taken and the degree of root galling was assessed using a modification of the method described by Daulton and Nausbaum (1961). The soil population of the juveniles was assessed as previously described. The storage roots and leaves of the harvested carrot plants were weighed, lengths of storage roots were measured and the soil population of juveniles assessed. Carrot was assessed for nematode-induced deformation of the main tap root as follows:

1. Main tap root not deformed.
2. Main tap root galled at the terminal portion only. Upper half not deformed. Galls mostly on lateral roots.
3. Main tap root completely deformed.

One-way analysis of variance was used to compare the effects of different treatments on the growth, root galling and soil population of the nematodes. Data obtained on fresh weight of leaves and root weight of lettuce and carrots respectively, were analysed using the two-way analysis of variance to test for interactions between the various factors - fertilizers, carbofuran and *M. incognita*.

RESULTS

LETTUCE

Based on fresh weight of leaves, the application of fertilizers and carbofuran alone or when applied together significantly ($P < 0.01$) increased top growth of infected lettuce plants (Table 3.4.1). Compared with uninfected plants given the same treatment, the top growth of infected plants in pots without fertilizers was reduced by 67.2%, but by 40.4% in pots to which fertilizers were added. When carbofuran was applied alone, there was a reduction of 22.3% but when applied with fertilizers a reduction of only 11.1% was found.

The growth of plants in pots given the combined application of fertilizers and carbofuran was not reduced by nematode infection ($P > 0.05$) (Table 3.4.1).

The interaction, fertilizers x nematode for top growth was not significant ($P > 0.05$) but carbofuran x nematode interaction was significant ($P < 0.01$). The interaction, fertilizers x nematicide for top weight was not significant ($P > 0.05$).

Root galling was reduced by the application of carbofuran alone or in combination with fertilizers. ($P < 0.05$), but was greater when carbofuran was applied together with fertilizers than when applied alone ($P < 0.05$) (Table 3.4.1).

The application of carbofuran alone to noninfested pots did not increase plant growth ($P > 0.05$)(Table 3.4.1).

The final soil population of juveniles was higher than the initial population of 1,200 juveniles, irrespective of treatment (Table 3.4.1). There was no difference between the final soil population found in pots to which carbofuran was applied alone or in combination with fertilizers.

CARROT

The weight of storage roots of infected plants was significantly ($P < 0.01$) increased by the application of fertilizers and carbofuran alone or when applied together (Table 3.4.2).

Compared with uninfected plants given the same treatment, the storage root weight of infected plants in pots without fertilizers was reduced by 78.7% but in pots given fertilizers only, a reduction of 42.5% was obtained. When carbofuran was applied alone there was a reduction of 39.4% but when applied together with fertilizers a reduction of 23.1% was obtained. Infected plants irrespective of treatment were significantly reduced by nematode infection ($P < 0.05$).

The interaction, fertilizer x nematode for storage root weights was not significant ($P > 0.05$), but the interaction, carbofuran x nematode was significant ($P < 0.01$). The interaction, fertilizer x nematicide was not significant ($P > 0.05$).

The length of storage roots of plants in pots without fertilizers was reduced by 62.7% but by 77.3% in pots with fertilizers. When carbofuran was applied alone root length was reduced by 39% but when applied with fertilizers root length was reduced by 26.2% (Table 3.4.2).

Table 3.4.1 Effects of fertilizers and carbofuran on the growth of lettuce infected with *M. incognita*

Treatments	Fresh weight of leaves (g) [1]	% Reduction in leaf weight	Fresh weight of roots	Gall index	No. of juveniles/ 200g of soil
Fertilizer + Nematodes	22.1	40.4	9.60	6.0	340.0
Fertilizer - Nematodes	37.6		8.40	0.0	0
Carbofuran + Nematodes	19.65	22.3	6.90	5.0	135.0
Carbofuran - Nematodes	25.30		6.10	0.0	0
Fertilizer + Carbofuran + Nematodes	34.20	11.1	10.10	5.5	152.5
Fertilizer + Carbofuran - Nematodes	38.45		9.20	0.0	0
No fertilizer No carbofuran + Nematodes	8.10	67.20	6.80	6.0	177.5
No fertilizer No carbofuran - Nematodes	24.73		6.07	0.0	0
L.S.D. at 5%	5.42		1.20	0.4	79.8
L.S.D. at 1%	6.54		6.45	0.6	111.9

[1] Means of four replicates

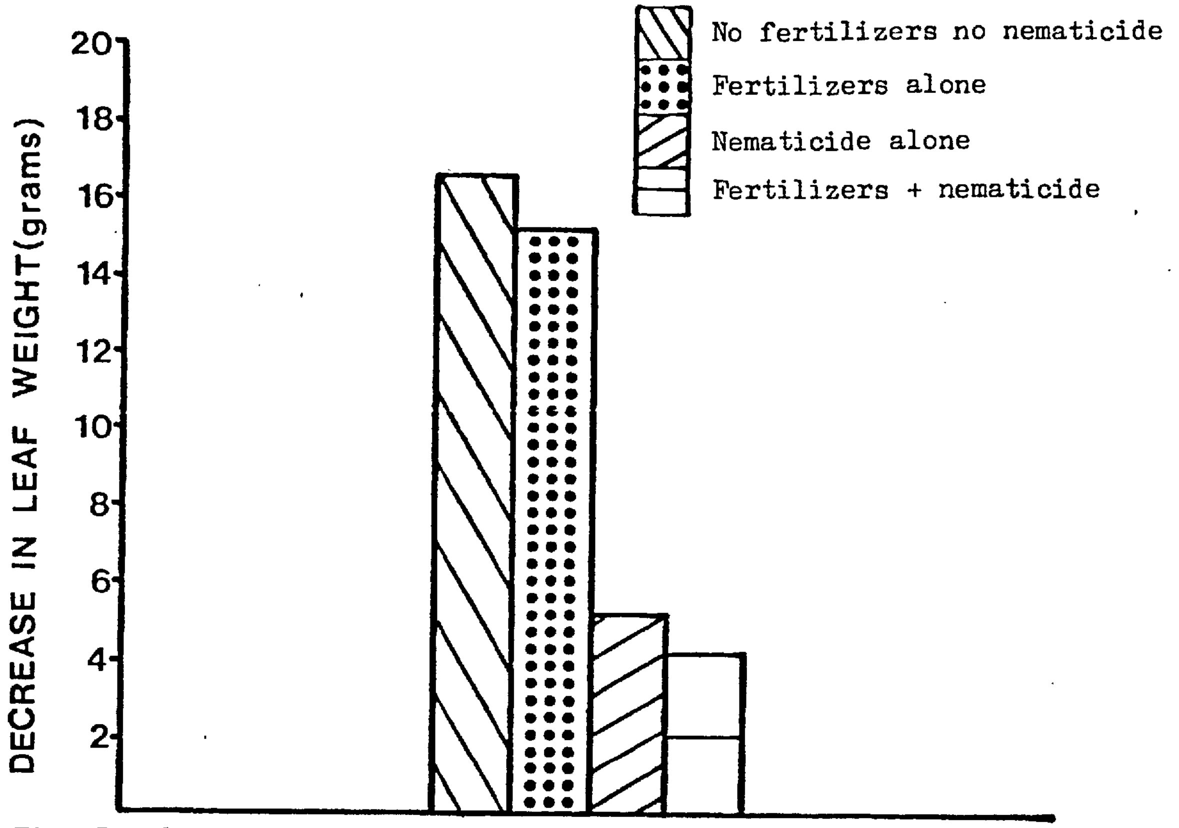


Fig. 3.3.1.

Reduction in leaf weight of infected lettuce plants
(Differences between the mean leaf weights of uninfected and infected plants).

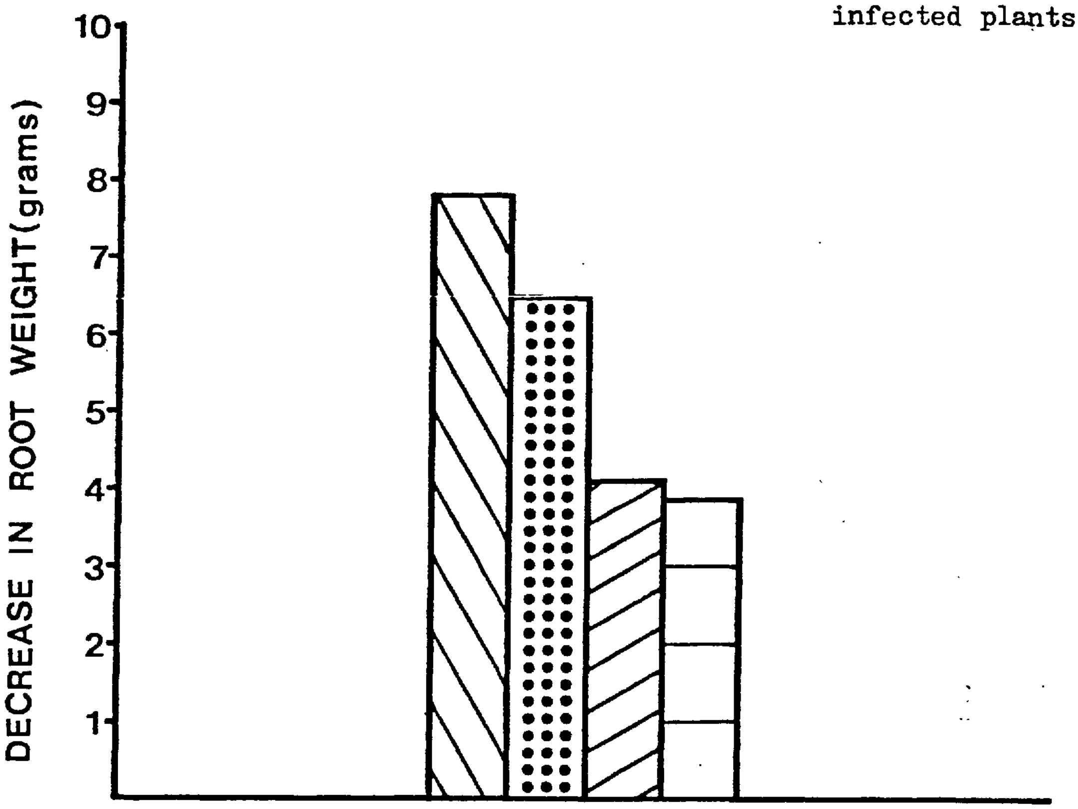


Fig. 3.3.2.

Reduction in storage root weights of infected carrot
(Differences between the mean root weights of uninfected and infected plants).

Table 3.4.2 Effects of fertilizers and carbofuran on the growth of carrot infected with *M. incognita*

Treatments	Fresh weight of storage roots (g) [1]	% Reduction in root weight	Root length in mm	Root deformation	Leaf weight (g)	No. of juveniles/ 200g of soil
Fertilizer + Nematodes	8.8	42.5	21.4	3.0	5.7	166.3
Fertilizer - Nematodes	15.3		94.2	0.0	7.9	0
Carbofuran + Nematodes	6.3	39.4	33.0	2.0	2.1	71.3
Carbofuran - Nematodes	10.4		54.1	0.0	2.9	0
Fertilizer + Carbofuran + Nematodes	13.3	23.1	71.8	2.0	6.2	85.0
Fertilizer + Carbofuran - Nematodes	17.3		96.0	0.0	7.1	0
No fertilizer No carbofuran + Nematodes	2.1	78.7	18.9	3.0	1.9	97.5
No Fertilizer No carbofuran - Nematodes	9.9		50.7	0.0	3.9	0
L.S.D. at 5%	2.1	-	8.6		1.5	47.3
L.S.D. at 1%	2.8		10.4		1.8	66.3

[1] Means of four replicates

Table 3.4.3 Effects of fertilizers on the growth of eggplant infected with *M. incognita*

Treatments	Fresh weight of tops (g) [1]	Fresh weight of roots (g)
Fertilizers + Nematodes	76.10	44.90
Fertilizers - Nematodes	86.20	38.50
No fertilizer + Nematodes	36.30	20.40
No fertilizer - Nematodes	58.78	24.85
L.S.D. at 5%	7.4	5.4
L.S.D. at 1%	10.4	7.6

[1] Mean of four replicates

Leaf weights were significantly ($P < 0.01$) reduced by nematode infection in pots given fertilizers, and in pots not given fertilizers, but leaf weight was not reduced when carbofuran was applied alone and when applied with fertilizers ($P > 0.05$) (Table 3.4.2).

Root deformation was reduced by the application of carbofuran alone or in combination with fertilizers (Table 3.4.2).

Final soil populations of juveniles were higher than the initial population of 1,200 juveniles, irrespective of treatment (Table 3.4.2). There was no significant difference between the final soil population found in pots to which carbofuran was applied alone or in combination with fertilizers ($P > 0.05$).

DISCUSSION

1. Effects of fertilizers and carbofuran on the growth of eggplant infected with *M. incognita*

Since the interaction between fertilizers and nematodes was significant (Table 3.3.1): the damaging effects of *M. incognita* on eggplant was markedly lessened following the application of fertilizers. Findings similar to this have been reported for the effects of fertilizers on the damaging effects of other plant parasific nematodes to crop plants. Ross (1969) found that soybean yields from microplots infested with *Heterodera glycines* were reduced by 71.8, 44.4 and 10.7% at the low, medium and high nitrogen levels respectively when compared with yields of non-infested plots at similar levels of N. Smolik and Malek (1973) reported that the pathogenic effect of *Tylenchorhynchus nudus* on Kentucky Blue grass was lessened through maintenance of adequate soil fertility. The good growth and yield of eggplant after the application of fertilizers (Table 3.2.2) even when infected with *M. incognita* was apparently due to increased root growth compensating for nematode root damage at their early stages of growth. This ensured that infected plants had enough healthy roots capable of absorbing sufficient nutrients from the soil. Since root infestation of these plants was not lessened when fertilizers were applied (Table 3.2.1 and Table 3.3.1), the tolerance of plants to damage by *M. incognita* was significantly improved.

Oteifa *et al.* (1965) also reported that increasing the amount of potassium alone or in combination with nitrogen and phosphorus seemed to induce a high degree of tolerance to cotton plants infected with

Tylenchorynchus latus. Vazquez (1967) found that plants given fertilizers tolerated higher infestations of *M. javanica* and suffered less damage than plants not given fertilizers.

The actual effectiveness of carbofuran application as a means of reducing crop losses due to *M. incognita* on eggplant has been shown (Table 3.3.1). Since carbofuran did not contribute through its stimulatory effect to increase plant growth (Table 3.3.1) the increased growth was due to the nematicidal action of carbofuran. However, the significant reduction in growth of infected plants despite the application of carbofuran is of practical significance, as it shows that nematodes recovering from the inhibitory effects of carbofuran can still cause significant crop losses.

The nematicidal action of carbofuran was not increased by the application of fertilizers. This occurred irrespective of time of application of fertilizers and carbofuran, as final soil populations of nematodes in soil given the combined application of fertilizers and nematicide were significantly higher than when carbofuran was applied alone (Table 3.2.2). Observations at six weeks after inoculation (Table 3.2.1) also showed more females per root system of plants in soil given fertilizers and treated with carbofuran than for plants in soil treated with only carbofuran. The nematicidal action of carbofuran was also not significantly reduced following the application of fertilizers. This is supported by the fact that the interaction between carbofuran and fertilizers was not significant (Table 3.3.1), indicating that the nematicidal action of carbofuran was the same when applied alone and in combination with fertilizers. The greater plant growth response to fertilizer application in soils treated with carbofuran (Table 3.2.2) indicates that carbofuran had no adverse effect on nutrient uptake at the different times of application. However, the greater reduction in nematode damage in terms of plant growth, when carbofuran and fertilizers were applied together (Table 3.3.1) shows they acted additively. This is of practical significance as it emphasizes the importance of correct fertilizer application even when nematicides are applied, as an additional means of reducing crop losses due to *Meloidogyne*.

The high incidence of root galling and the very high final soil populations of nematodes in treated soil (Table 3.2.2) indicate that the nematicidal effect of carbofuran on the nematodes was of short duration. The nematode population in treated soil equalled those in the untreated, probably because the roots of plants in treated soil were healthier and more extensive, and therefore more favourable for reproduction when the nematodes recovered from the effect of carbofuran. The very short life cycle of *M. incognita* may also have contributed to the high increase. Kinlock (1974) also found that the final soil populations under different soybean cultivars in soil treated with carbofuran were equal to that of the untreated at harvesting, even though yields were significantly increased. Final soil populations in treated soil which were as high as that of the untreated have also been reported by several workers for the control of *H. schachtii* on beet with aldicarb (Steudel and Thieleman, 1967, Jorgenson, 1969, Thielemann and Steudel, 1973).

The high final populations in soils given fertilizers alone were related to root development. Hesling (1959) and Oostenbrink (1960) noted that fertilizers and organic manures could improve root development which in turn could promote populations of plant-parasitic nematodes. Haque *et al.* (1972) attributed faster development of *M. incognita* on okra roots to the higher root weights which provided more feeding sites for the nematodes.

2. Effects of fertilizers and carbofuran on the growth of lettuce and carrot infected with *M. incognita*

The enhanced growth of infected lettuce and carrot plants (Table 3.4.1 and 3.4.2), following the application of fertilizers shows that these plants were able to utilize some of the nutrients in the soil. Since the interaction between fertilizers and nematodes was not significant for leaf and storage root weights of lettuce and carrot respectively, the damaging effect of *M. incognita* on these crops was not influenced by fertilizer application. The damage caused by *M. incognita* to these crops, in terms of crop losses, was unaltered by the application of fertilizers when compared to the losses suffered by plants in soil without fertilizers (Figures 3.3.1 and 3.3.2). This result is similar to that obtained by Trudgill *et al.* (1975) who reported that, although overall yield of potatoes infected by *Globodera rostochiensis* was increased by the application of NPK fertilizers, the proportion of yield loss was not reduced. But the damaging effects of *M. incognita* on eggplant, grown under similar conditions as lettuce and carrot, and inoculated with the same number of nematodes, were significantly lessened by fertilizer application (Table 3.4.3). Lettuce and carrot probably lack the capacity to compensate adequately for loss of functional roots due to nematode attack. According to Jones (1977), in pest-crop situations transplants are less vulnerable because they represent more plant material than those grown directly from seed and so in establishing root and shoot systems, are better able to replace roots or buds that are destroyed. However, the significant increase in the growth of infected lettuce and carrot shows that despite apparently early attack by nematodes, they compensated to some extent for damaged roots. This supports the view that plants respond to the attack of nematodes by attempting to regenerate and repair the damaged roots (Wallace, 1973).

The significant interaction between carbofuran and nematodes for the fresh weight of leaves and storage roots of carrots respectively (Table 3.4.1 and 3.4.2) shows that carbofuran significantly reduced crop losses due to *M. incognita*. Since carbofuran applied at the rate used did not stimulate the growth of these plants (Table 3.4.1 and 3.4.2) the increase in plant growth of infected plants was due to its nematicidal action. The results obtained for carrot (Table 3.4.2) show that the plants were still significantly damaged even when carbofuran and fertilizers were applied. This may be related to its slow development during the early stages of growth. The slow growing plants could not escape severe damage after the recovery of the nematodes from the effect of carbofuran. In contrast, lettuce completely escaped nematode damage when carbofuran and fertilizers were applied, whereas plants in soil treated with only carbofuran still suffered significant losses (Table 3.4.1). A possible explanation may be that plants in soil treated with carbofuran and given fertilizers developed a larger root system which helped to reduce the stress of nematode injury when the nematodes recovered from the effect of carbofuran.

Since the interaction between fertilizers and nematicide was not significant for leaf weight and storage root weights of lettuce and carrots respectively (Table 3.4.1 and 3.4.2), the effect of carbofuran on damage caused by *M. incognita* to the plants was the same when applied alone or in combination with fertilizers. This is supported by the similar final soil populations found when carbofuran was applied alone or in combination with fertilizers (Table 3.4.1 and 3.4.2). The high final soil populations found at harvesting indicates that carbofuran was of short persistence and that high final soil populations should be expected when it is applied in the production of these crops under similar conditions.

SECTION 4

MECHANISM OF ACTION OF FERTILIZERS ON *M. INCOGNITA* INFECTING EGGPLANT.

INTRODUCTION

Although the results from previous experiments (Section 3.2) showed that NPK fertilizers lessened the damaging effects of *M. incognita* on eggplant, the mechanism of action could not be properly ascertained.

Jones (1977) lists several ways by which fertilizers may influence the relationship between pests in general including nematodes and crop plants in favour of the crop. Fertilizers may enable plants to escape severe damage through enhanced growth so that the period of activity of the pest is not properly synchronized with the vulnerable stages of the host. They could improve the tolerance of the host because it is able to replace damaged parts by compensatory growth or by failing to react to virulent toxins which in susceptible plants, cause necrotic lesions (hypersensitivity). Some hosts may be less heavily attacked or avoided because of a deficiency in the strain of stimuli that leads to feeding or oviposition or they contain substances that deter or repel. This is usually referred to as nonpreference. Antibiosis can also occur when the quality of food is inappropriate, nutrients are in the wrong proportions, the food is unpalatable, sticky hairs in the case of insects or other morphological impediments are present; or there is an excess of fibres or harmful toxins that interfere with growth and reproduction.

Fertilizers may also influence nematodes in the soil directly; for example addition of 70 and 700ppm of nitrogen to soil decreased populations of *P. penetrans* (Walker, 1971). In other studies, Barker *et al.* (1971) showed that the application of $NaNO_3$ and NH_4NO_3 to soil decreased hatch, penetration and cyst development of *Heterodera glycines* on soybeans. Oteifa (1955) found that ammonium - N, when compared to nitrate -N, inhibited egg hatch and initial penetration of juveniles into roots of lima beans, but had no effect on subsequent rate of nematode development and reproduction after penetration.

Application of high rates of fertilizers may cause a significant increase in the osmotic pressure of soil water which could in turn inhibit nematodes (Wallace, 1971a). Soil pH may also be changed by the application of fertilizers; there is evidence that pH may be inhibitory to *Meloidogyne javanica* below 5.0 and above 8.0 (Wallace, 1966).

Against the above background, the following laboratory and glasshouse experiments were carried out to determine the possible mechanism by which NPK fertilizers used in previous studies, lessened the detrimental effects of *M. incognita* on eggplant.

4.1 INFLUENCE OF FERTILIZERS ON ROOT INVASION

MATERIALS AND METHODS

Plastic pots, 22.5cm, in diameter, were each filled with 5kg of heat sterilized soil. Fertilizers were added at the rate used in previous experiments (18.29 grams/pot), 1.5X and 2X this rate and none. The pH of the soil was determined before and after the addition of fertilizers (see General Materials and Methods).

Four-week old seedlings of eggplant were transplanted singly into each pot with four replicates per fertilizer rate. These plants were each inoculated with 500 second stage juveniles of *M. incognita* and the pots were arranged in a fully randomized design on a glasshouse bench where the temperature ranged from 17^{o} - 38^{o}C.

Fourteen days after inoculation, root systems were carefully removed from each pot and washed free of adhering soil particles. Whole root systems were macerated in a Waring Blender after staining in boiling lactophenol containing 1% (w/v) cotton blue. The macerated roots were poured into a beaker and made up to $100cm^3$. Estimates of the number of nematodes per root system were determined from three $10cm^3$ aliquots taken from the suspension.

RESULTS

The results are summarized in Figure 4.3.1. Invasion by second stage juveniles was not significantly reduced by the application of fertilizers at the normal rate (18.29g per pot) and 1.5 X the normal rate ($p > 0.05$) but 2 X the normal rate did significantly reduce root invasion ($p < 0.01$).

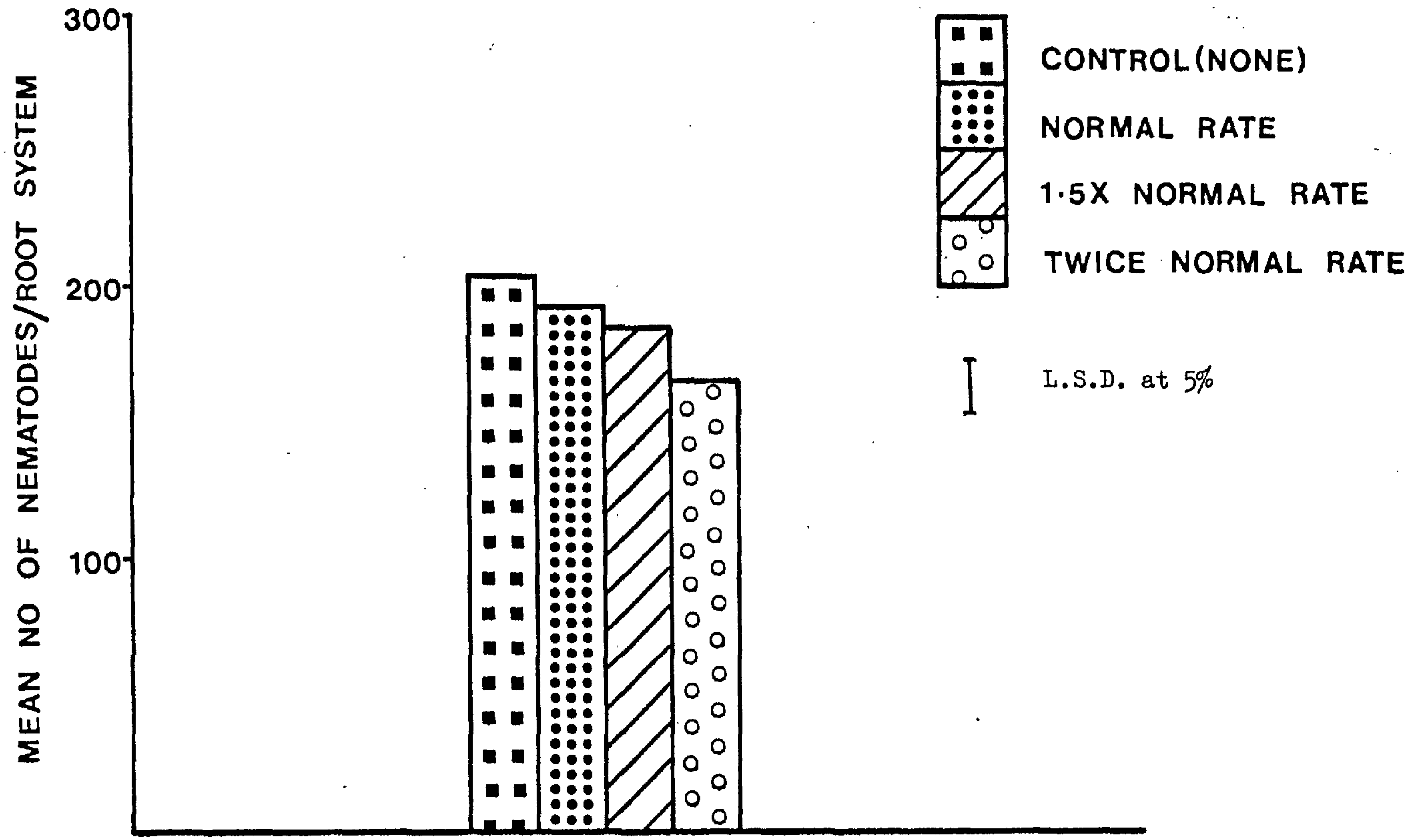


FIG .4.3.1 Influence of different rates of fertilizers on root invasion by *M.incognita*. Each value is a mean of four replicates.

4.2 INFLUENCE OF FERTILIZERS ON SHOOT AND ROOT DEVELOPMENT OF EGGPLANT INFECTED WITH *M. INCOGNITA*

MATERIALS AND METHODS

Forty-eight, 22.5cm plastic pots were each filled with 5kg of heat sterilized soil. Fertilizers were added to half of these pots at the rate of 18.29g per pot, while the other half were not given fertilizers. Four-week old eggplant seedlings of uniform size were transplanted singly into each pot. Half of the pots given fertilizers were inoculated at the rate of 2,000 freshly hatched second stage juveniles per pot, while the other half were left uninoculated. Pots without fertilizers were inoculated in a similar manner. Details of the various treatments are given in Table 4.3.1.

Pots were arranged in a fully randomized design in the glasshouse where the temperature ranged from 16^{o} - 40^{o}C.

At three and six weeks after inoculation, four inoculated and four uninoculated plants in fertilized soil were harvested. A similar number of inoculated and uninoculated plants in unfertilized soil were also harvested.

Data were taken on fresh and dry shoot weight, fresh weight of roots, length of roots, number of galls per gram of root and per root system. The proportion of roots galled was assessed as follows:-

1 - up to 20% of roots galled.

2 - 30 - 40% of roots galled.

3 - 50 - 60% of roots galled.

4 - 70 - 80% of roots galled.

5 - 90 - 100% of roots galled.

Six weeks after inoculation, fertilizers were added to the remaining pots which were given fertilizers at the start of the experiment at the rate of 9.14g per pot.

At nine weeks after inoculation, the experiment was terminated. Unfortunately, the number of galls per root system could not be properly assessed because of the presence of many coalesced galls. The shoot weight was also not recorded because of the attack of red spider mites which caused large difference between replicates. Data were therefore taken only on the proportion of roots galled, root weight and length.

RESULTS

At three weeks after inoculation, the fresh and dry shoot weights of infected plants in fertilized soil were significantly greater than those of infected plants in soil not given additonal fertilizers ($p<0.01$) (Table 4.3.1). Similarly the fresh and dry shoot weights of uninfected plants were significantly greater than the corresponding ones in unfertilized soil ($p < 0.01$) but no significant differences were found between the shoot weights of infected and uninfected plants in both soils ($p > 0.05$). By six weeks after inoculation, the fresh and dry shoot weights of uninfected plants were significantly greater than those of infected plants ($p < 0.01$). This applied to plants in fertilized and unfertilized soil. Between the third and sixth week after inoculation, the mean fresh and dry shoot weights of infected plants in fertilized soil increased by 33.3g and 5.58g respectively (Table 4.3.1). During the same period the mean fresh and dry weights of uninfected plants increased by 44.5g and 7.73g respectively. But in soil not given additional fertilizers, the mean fresh and dry shoot weights of infected plants increased by 4.3g and 0.96g respectively, while the fresh weights of

Table 4.3.1. <u>Effects of fertilizers on shoot development</u>

Fresh and dry shoot weights

	Time after inoculation			
	3 weeks		6 weeks	
Treatments	Fresh shoot weight 1	Dry shoot weight	Fresh shoot weight	Dry shoot weight
Fertilizer + Nematodes	26.4	4.39	59.7	9.97
Fertilizer − Nematodes	30.3	4.92	74.8	12.65
None + Nematodes	13.9	2.36	18.2	3.02
None − Nematodes	19.1	3.12	49.3	8.23
L.S.D. at 5%	5.88	0.96	7.12	1.20
L.S.D. at 1%	8.25	1.34	9.99	1.68

1 Means of 4 replicates

Increase in root weight with time

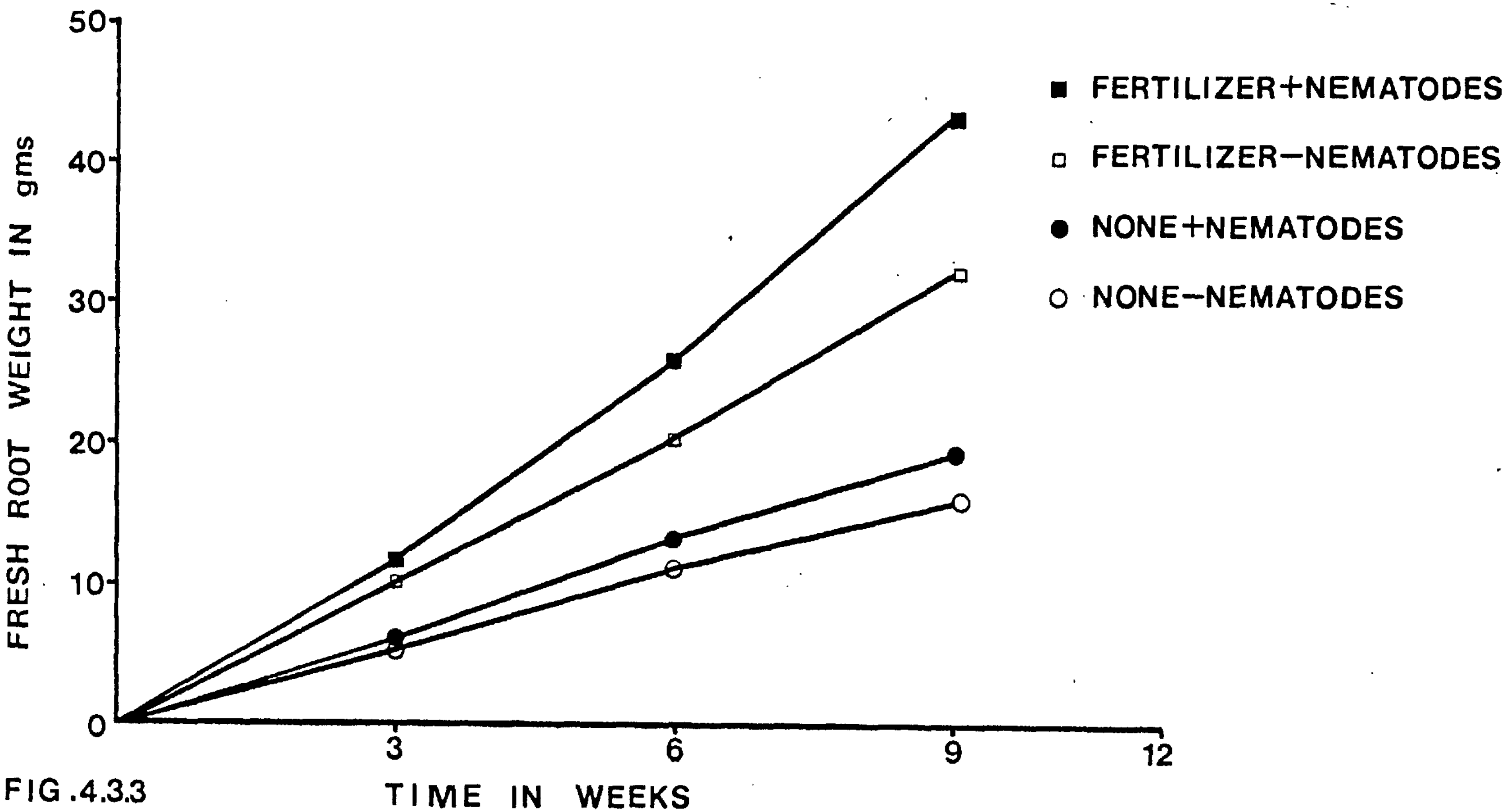


FIG.4.3.3

Increase in root length with time

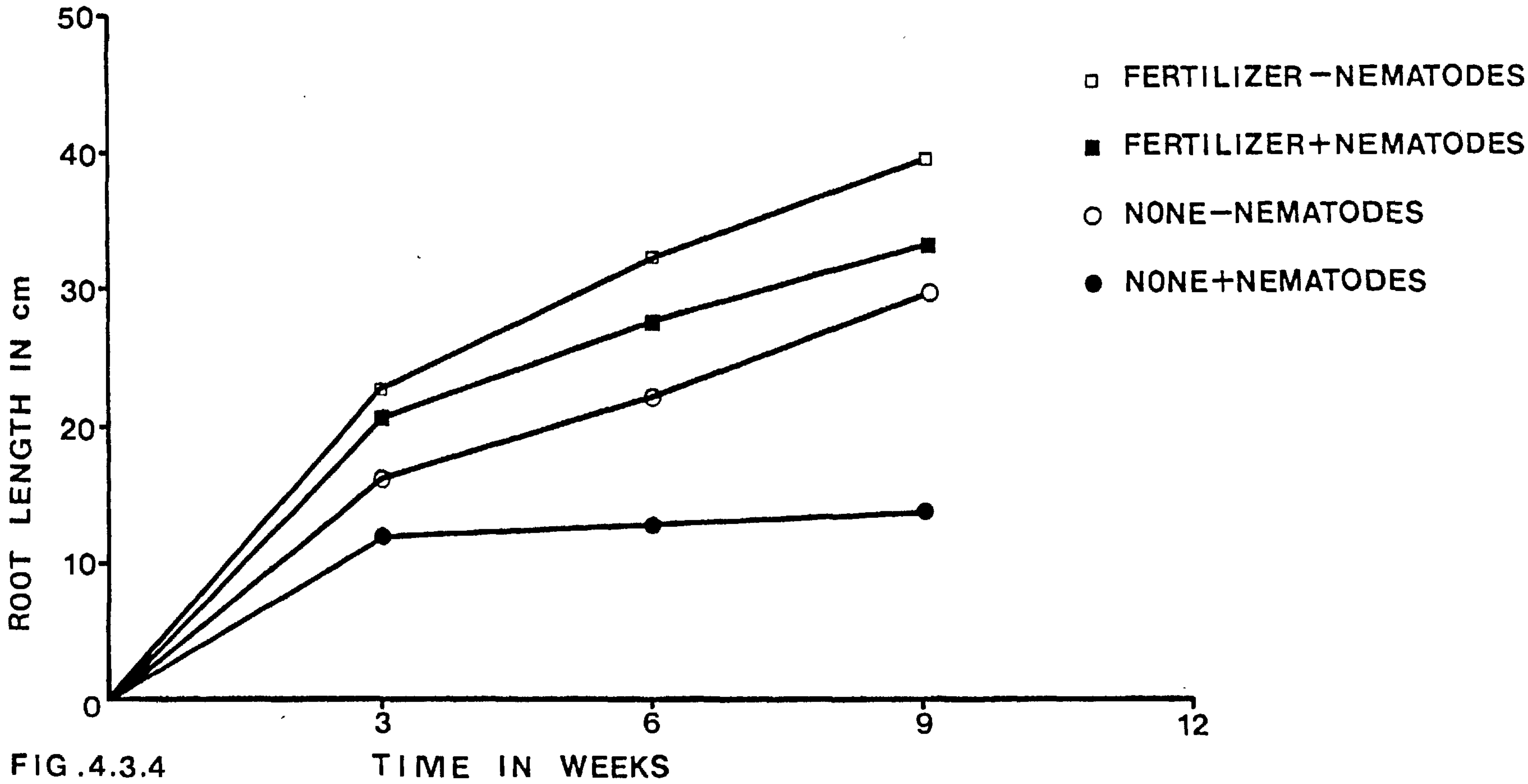


FIG.4.3.4

Increase in proportion of roots galled with time

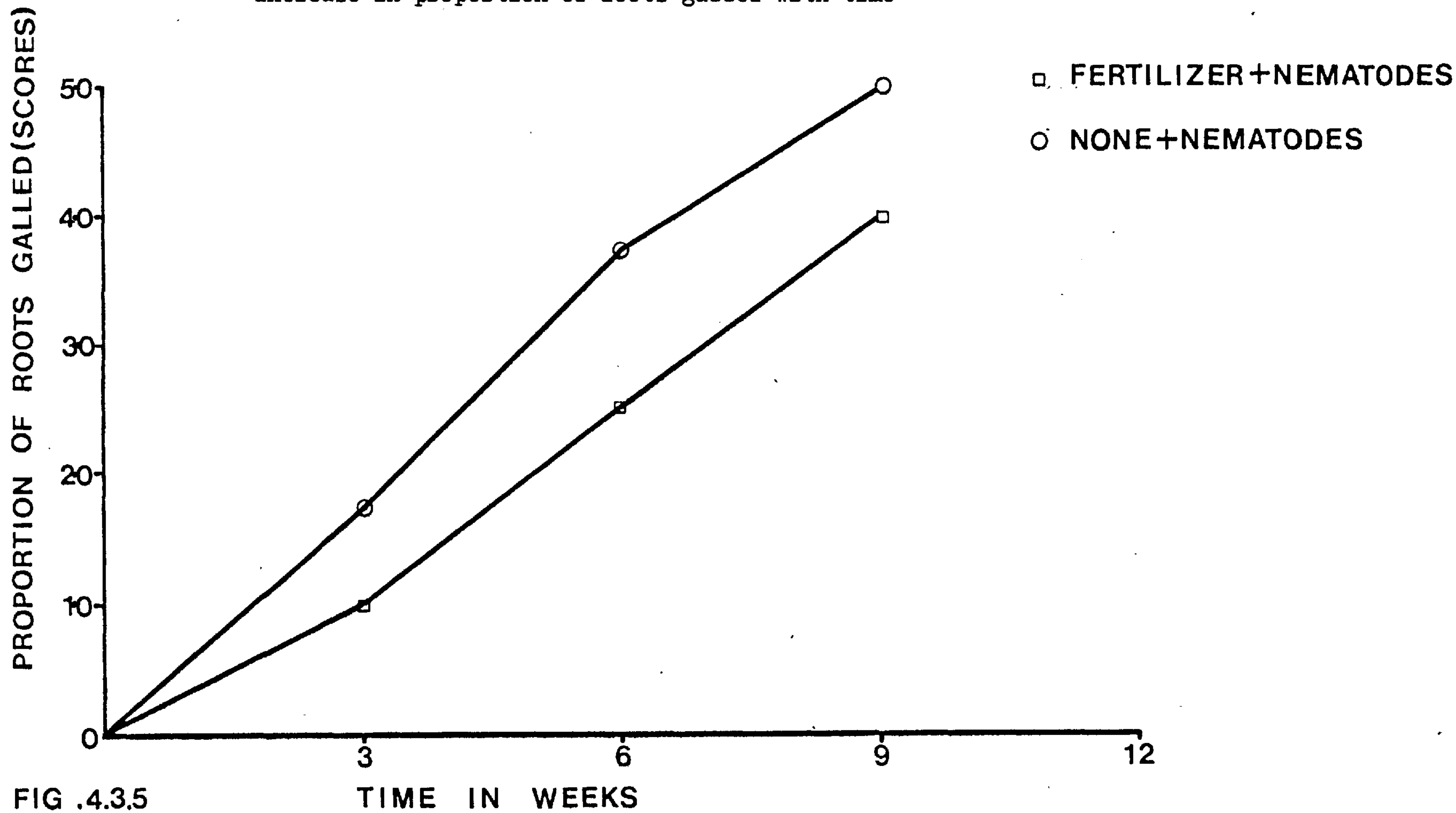


FIG .4.3.5

Table 4.3.2 Effects of fertilizers on root development.

Time after inoculation

Treatments	3 weeks		6 weeks		9 weeks	
	Fresh weight of roots (g)1	Root length in cm	Fresh weight of roots (g)	Root length in cm	Fresh weight of roots (g)	Root length in cm
Fertilizers + Nematodes	12.8	20.4	26.7	27.9	43.0	33.2
Fertilizers - Nematodes	10.1	22.8	20.5	32.4	32.4	39.6
None + Nematodes	6.4	12.8	13.2	13.9	18.9	14.4
None - Nematodes	5.7	16.9	11.6	22.4	16.4	29.6
L.S.D. at 5%	1.9	5.7	4.0	6.6	5.5	
L.S.D. at 1%	2.6	7.9	5.6	9.3	8.1	

1 Means of 4 replicates

uninfected plants increased by 30.1g and 5.1g.

At three weeks after inoculation, the root weights of infected and uninfected plants in soil given fertilizers were significantly greater than the root weights of corresponding plants in soil without fertilizers ($p < 0.01$) (Table 4.3.2). The root weights of infected plants in soil given fertilizers were significantly greater than those of uninfected plants ($p < 0.01$), but in soil without fertilizers there was no significant difference between the root weights of infected and uninfected plants ($p > 0.05$). The root weights and lengths of infected and uninfected plants in both soils increased with time (Figure 4.3.3 and Figure 4.3.4). The increase in root weight with time was always greater for infected plants than uninfected plants. The rate of increase in root weight and length was also greater in soils given fertilizers than in unfertilized soil. The rate of increase in root length was greater for uninfected plants (Figure 4.3.4).

Between the third and sixth week, the root length of infected plants in fertilized soil increased by 7.5cm while those of uninfected plants increased by 9.6cm but in unfertilized soil the increases were 1.1 and 5.5cm for infected and uninfected plants respectively. Between the sixth and ninth week, the root length of infected and uninfected plants in fertilized soil increased by 5.3cm and 7.2cm respectively but in contrast the increases in root length for infected and uninfected plants were 0.5cm and 7.2cm respectively.

The number of galls per root system and per gram of root increased with time in fertilized soil and soil without fertilizers (Figures 4.3.6 and 4.3.7). No significant ($p > 0.05$) difference was found between the number of galls per root system in both soils, three weeks after

Increase in number of galls per root system with time

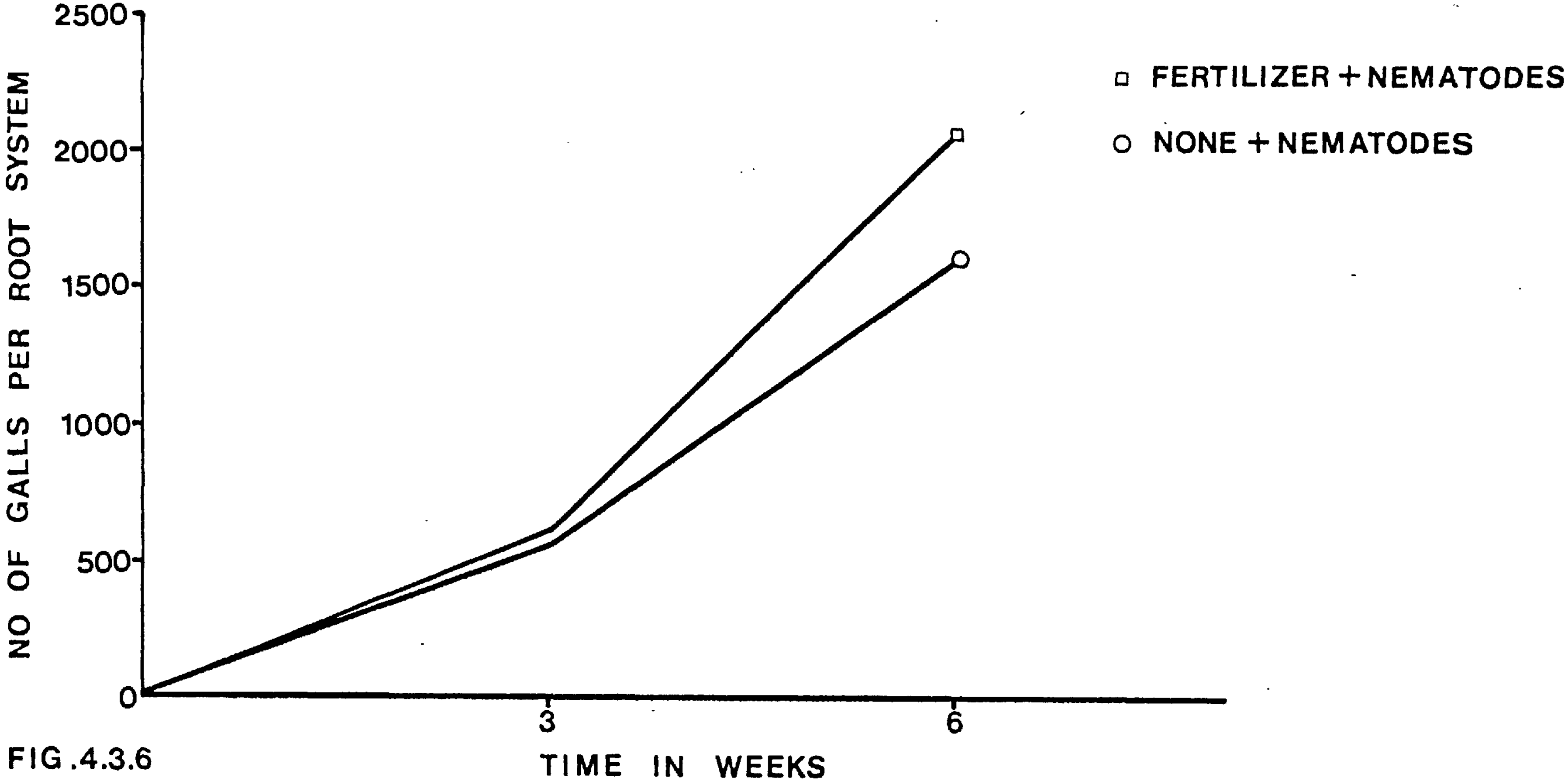


FIG.4.3.6

Increase in number of galls per gram of root with time

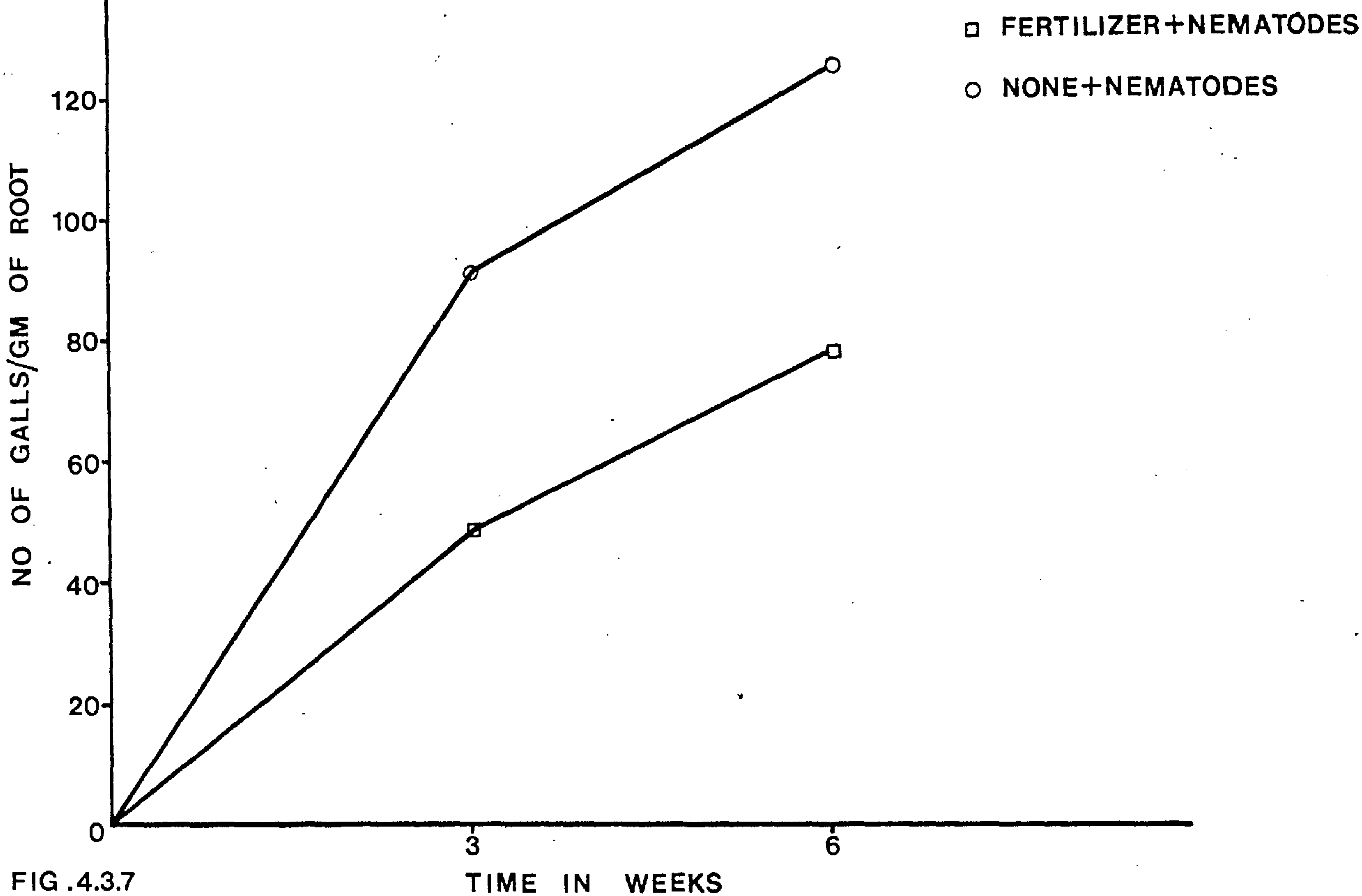


FIG.4.3.7

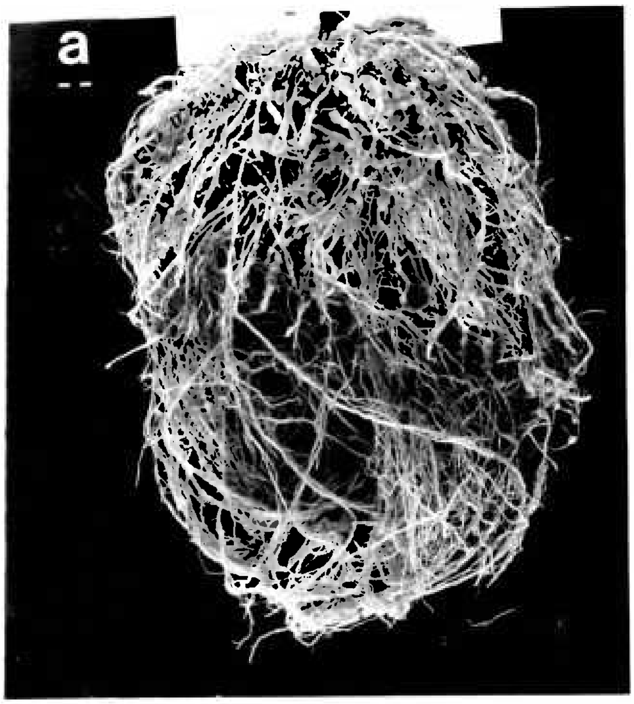
inoculation, but the number of galls per gram of root in unfertilized soil was significantly greater than was obtained for plants in soils given fertilizers ($p < 0.05$). At six weeks after inoculation, the number of galls per root system was significantly greater for plants in soil given fertilizers ($p < 0.05$), but the number of galls per gram of root was significantly less ($p < 0.05$) (Figure 4.3.7).

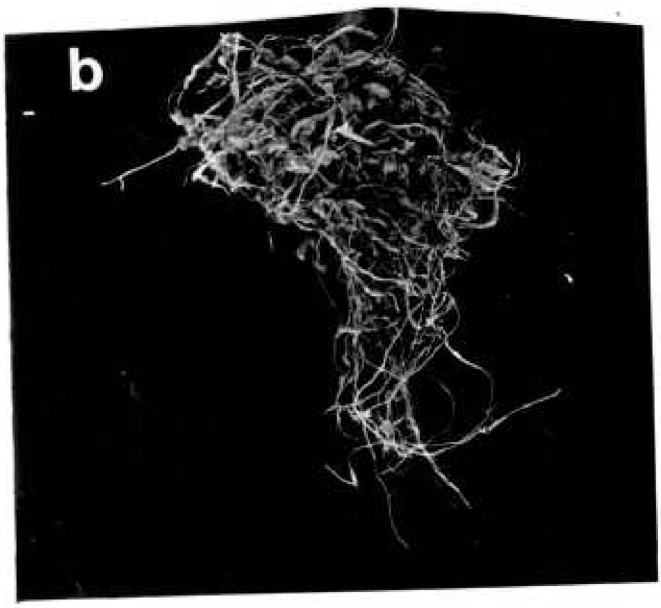
The proportion of roots galled increased with time until nine weeks after inoculation, when the experiment was terminated (Figure 4.3.5). This trend was found for infected plants in soils given fertilizers and those in unfertilized soil. At three, six and nine weeks after inoculation, a greater proportion of the roots of plants in soil not given fertilizers was galled when compared with those of plants in fertilized soil. At the termination of the experiment, nearly all roots of plants in unfertilized soil were galled. In contrast, up to 20% of the roots of plants in fertilized soil were free from galls.

a. Galled root system of eggplant in pots given fertilizers. (63 days after inoculation).

b. Galled root system of eggplant in pots without fertilizers.

(63 days after inoculation).


a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4.3 *IN VITRO* EFFECTS OF DIFFERENT CONCENTRATIONS OF FERTILIZER SUSPENSION ON SECOND STAGE JUVENILES OF *M. INCOGNITA*

MATERIALS AND METHODS

A suspension of 10,000ppm of the NPK fertilizer was prepared by dissolving 10grams in distilled water. By serial dilution, concentrations of 1,000, 100 and 10ppm were prepared. A buffered series (Na_2HPO_4 - KH_2PO_4) of fertilizer solutions at pH7 was included for comparison.

Ten freshly hatched second stage juveniles were deposited into syracuse dishes containing $2cm^3$ of the different concentrations of the fertilizer. Nematodes in distilled water served as control. There were three replicates for each concentration. All the dishes were covered with glass plates and placed in an incubator held at 25^oC. The activity and mortality of the nematodes was checked hourly for the first six hours and every 24 hrs for the next four days. Final assessment was made one week after the commencement of the experiment. Nematodes were considered inactive if they moved only when touched with a needle; those that did not respond to touch were considered as dead.

RESULTS

Fertilizers at 10ppm and 100ppm had little effect on the activity and survival of the nematodes (Table 4.4.1). At 24 hrs only 50 and 36% of the nematodes at 1000ppm and 10,000ppm respectively were active. There was no nematode mortality up to 96 hours but at 7 days, 40% of the nematodes at 10,000ppm were dead. Nematodes at 1000ppm still responded to touch at 7 days.

The survival of the nematodes in the buffered series was the same (Table 4.4.2).

Table 4.4.1. Survival of second stage juveniles of *M. incognita* in fertilizer solutions

Percentage active nematodes

	Concentration [1]				
Time (hrs)	0	10ppm	100ppm	1000ppm	10,000ppm
6	100	100	100	100	100
24	100	100	100	50	36
48	100	100	100	40	23
72	100	100	76	30	13
96	100	100	73	23	0
7 days	83	73	63	16	0

1) Means of 3 replicates.

Table 4.4.2 Survival of second stage juveniles of *M. incognita* in fertilizer solution (Buffered series)

Percentage active nematodes. 1)

	Concentration				
Time (hrs)	0	10ppm	100ppm	1000ppm	10,000ppm
6	100	100	100	100	100
24	100	100	100	53	40
48	100	100	100	40	20
72	100	100	80	33	10
96	100	100	70	30	0
7 days	83	73	66	23	0

1) Means of 3 replicates

DISCUSSION

The results obtained from the experiment on root invasion (Figure 4.3.1) showed that fertilizers applied at the normal rate had no inhibitory effect on the nematodes; their role was purely nutritional. However, the reduction in root invasion at twice the normal rate of fertilizer application is of practical significance, as it suggests that the use of very high rates of this fertilizer might be of benefit, provided phytotoxicity can be avoided. As the application of fertilizers at this rate did not significantly change soil pH, fertilizers appeared to affect the nematodes directly. This is supported by the loss of activity by the nematodes immersed in high concentrations of fertilizer *in vitro* (Table 4.4.1). The nitrogen component of this fertilizer may have been responsible, as the detrimental effect of ammonium containing compounds to nematodes has been reported by several workers (Oteifa, 1955; Walker, 1971; Barker *et al.*,1971, Mankau and Mankau, 1975).

In the experiment on shoot and root development, the results obtained for shoot weights at six weeks, showed that there was a greater difference between the growth rates of infected and uninfected plants grown in soil without additional fertilizers (Table 4.3.1). This infers that nutrient uptake by the roots of infected plants was more seriously impaired in soil without additional fertilizers. The marked increase in shoot weight of infected plants growing in soil given fertilizers between the third and sixth week (Table 4.3.1) shows that plants were able to utilize available nutrients in the soil, though less efficiently than uninfected plants. This was probably due to the presence of many roots which were free from galls. The healthy roots represent new ones generated to compensate for damaged roots as plants attacked by nematodes often attempt to offset the damage by root proliferation (Malek and Jenkins, 1964; Madamba *et al.*, 1965), brought about through hormone regulation.

The greater proportion of roots not galled for plants in soil given fertilizers is therefore due to the greater capacity of these plants to compensate for damaged roots. The ability of nematode-infected plants to compensate for damaged roots appears to be related to the external concentration of nutrients around the roots. The observed localized proliferation of barley roots in the area enriched with nutrients (Drew *et al.*, 1973; Drew and Saker, 1975), seems to show this. It was evident from the results of their experiment that increased availability of nutrients to the roots caused them to receive an enhanced fraction of the metabolites which are translocated from the shoots.

The results obtained on root-galling (Figures 4.3.6 and 4.3.7) and shoot development (Table 4.3.1) showed a correlation between the number of galls per gram of root and the reduction in plant growth due to the nematodes. Plants in fertilized soil which had more galls per root system but fewer galls per gram of root were less affected by the nematodes. The fewer galls per gram of root therefore shows a dilution of intensity of attack per unit of root weight, due to the effect of fertilizers on root growth. Johnson (1971) also related fewer numbers of galls per gram of root to reduction in severity of root-knot damage.

The positive correlation between the proportion of root system not galled and shoot growth is in agreement with the hypothesis of Seinhorst (1965). He considered that the amount of damage caused by a nematode is related to the preplant population density of the nematodes and the amount of exposed tissue not attacked. It is suggested that the estimation of the proportion of roots not galled, at the early stages of the growth of infected plants might be useful in predicting subsequent crop losses.

A thorough appraisal of all the results obtained from the above experiments, show that the damaging effect of *M. incognita* was lessened in fertilized soil because of their enhanced capacity to compensate for damaged roots during the early stages of growth. Since root infestation was not reduced by the application of fertilizers, it implies that fertilizers improved the tolerance of the plants to the damaging effects of *M. incognita* in a significant way.

SECTION 5

MECHANISM OF ACTION OF CARBOFURAN ON *M. INCOGNITA*

INTRODUCTION

The nematicidal efficacy of carbofuran against *M. incognita* was first reported by Di Sanzo (1969) and several similar reports have appeared since then (Kinlock 1974 ; Reddy and Seshadri, 1971; Sivakumar *et al.*, 1974; 1976; Overman&Jones,1975).

The earliest attempt to determine the mode of action of carbofuran against *M. incognita* was by Di Sanzo in 1969. He found that roots of tomato plants grown in soil treated with carbofuran at 10ppm, were free from galls compared to the heavily galled roots of plants in untreated soil. Greater numbers of nematodes were recovered from treated than untreated soil, suggesting that carbofuran was not lethal at the rate applied but prevented the nematodes from invading the roots. Second-stage juveniles of *M. incognita* placed in petri dishes containing 10ppm carbofuran in nutrient agar medium moved at random and did not infect tomato seedlings germinated in the dishes. Nematodes deposited in untreated medium moved towards the roots and established infection. From further studies by Di Sanzo (1973) using *Pratylenchus penetrans* and *Tylenchorhynchus claytoni* he suggested that carbofuran affects plant-parasitic nematodes by altering their orientation and feeding mechanisms.

The question of whether or not carbofuran may act systemically to control nematodes within roots of host plants has not been satisfactorily answered. Reddy and Seshadri (1971) found that the application of carbofuran one week after inoculation reduced the number of galls on tomato roots. Sivakumar *et al.* (1974, 1976) reported increased yields of tomato grown in soil infested with *M. incognita* when carbofuran was applied ten days after transplanting. Results from the experiment reported in Section 3.1 showed that the number of females in the roots of eggplant grown in soil treated with carbofuran two weeks after transplanting was less than those found in the roots of plants in untreated soil. Since it was not clear if the observed effects discussed above are due to systemic action of carbofuran against the nematodes already in the roots; an investigation was needed to elucidate this.

There are several other aspects of the interactions between plants, *M. incognita* and carbofuran which could help to throw more light on the mechanism by which carbofuran controls *M. incognita*.

These include the following:-

(i) the possible role of the metabolites of carbofuran in the control of *M. incognita*.

(ii) the immediate and residual effects of carbofuran on egg hatch.

(iii) how concentration and duration of exposure of second stage juveniles to carbofuran affect their invasion.

(iv) the effect of nematicides in the roots on the control of soil nematodes.

(v) the effect of carbofuran on migration.

(vi) the behavioural responses of the second stage juveniles in treated medium in the presence of a host plant.

Against the above background, a series of integrated laboratory and glasshouse experiments were done to work out the mechanism by which carbofuran controls *M. incognita* at field rates.

5.1 EFFECTS OF CARBOFURAN AND 3-HYDROXYCARBOFURAN ON POSTURE, ACTIVITY AND ROOT INVASION BY *M. INCOGNITA*

Studies carried out on the degradation of carbofuran in soils and plants have identified 3-hydroxycarbofuran and 3-ketocarbofuran as the two common metabolites encountered (Caro *et al.*, 1973; Turner and Caro, 1973; Talekar *et al.*, 1977). The possible role of these metabolites in the control of *M. incognita* had not been reported.

As 3-hydroxycarbofuran was available, it was decided to assess its importance in the control of *M. incognita* by comparing its effects with that of carbofuran, on posture, activity and root invasion.

An experiment was also set up to determine if the invasive power of the juveniles is affected after exposure to different concentrations of carbofuran for different times.

MATERIALS AND METHODS

1. Posture and activity

Concentrations of 1, 2, 5 and 10ppm of carbofuran and 3-hydroxycarbofuran were prepared from their stock solutions by serial dilution, and 1% (v/v) acetone was used as the control. Each treatment was replicated three times. The dishes were covered with small glass plates and kept in an incubator held at 25°C. The nematodes were examined for posture and activity at 2, 4, 6, 24 and 48 hours. The nematodes were rated for change of posture as shown in Figure 5.2.1. The activity of the juveniles was based on body movement and a scale of 1 - 6 was used to rate the activity of the nematodes as follows:

1 - no movement of juveniles within 10 seconds.

2 - juveniles moved within 8 - 10 seconds.

3 - juveniles moved within 5 - 7 seconds.

4 - juveniles moved within 2 - 4 seconds.

5 - movement normal and similar to that of control.

6 - hyperactivity observed.

At the end of the 48 hours observation period, the nematodes were transferred into syracuse dishes containing distilled water. Their activity was recorded after 6 hours to check for recovery.

2. Effect of carbofuran and 3-hydroxycarbofuran on invasion of tomato roots by *M. incognita*

This experiment was carried out in a constant environment chamber, with humidity and temperature maintained at 75% and 25°C respectively and a 14 hour day light period.

Small glass vials, $10cm^3$ in volume, were each filled with 5g of washed dried sand (<180μ). $1cm^3$ suspension of 350 juveniles in 1% (w/v) phostrogen solution were pipetted into each glass vial. $1cm^3$ of the pesticide solutions dissolved in 1% (v/v) acetone was also pipetted into each vial. After incubation at 25°C for 24 hours, a tomato seedling (var. Moneymaker) at the two-leaf stage was taken, its roots removed under water with a sharp scalpel and the cut stem placed a few mm into the moist sand. This is the PI test of Bunt (1975). Ten days later, the plants were carefully removed from the vials and the newly produced roots washed thoroughly, stained in 1% (w/v) cotton blue in lactophenol. The roots were then examined under a dissecting microscope to determine the number of nematodes in the roots.

Details of the treatments were as follows:-

Control I (1% (w/v) phostrogen solution only.

Control II 1% (v/v) acetone only.

$1cm^3$ of 10ppm carbofuran (2μg/g of sand)

$1cm^3$ of 20ppm carbofuran (4μg/g of sand)

$1cm^3$ of 40ppm carbofuran (8μg/g of sand)

$1cm^3$ of 10ppm 3-hydroxycarbofuran (2μg/g of sand)

$1cm^3$ of 20ppm 3-hydroxycarbofuran (4μg/g of sand)

$1cm^3$ of 40ppm 3-hydroxycarbofuran (8μg/g of sand)

Each of the above treatments were replicated four times.

3. <u>Invasion of second stage juveniles after exposure to different concentrations of carbofuran for different periods</u>.

Two cm^3 of carbofuran solutions (4, 8 or 16ppm) or control solution were pipetted in test tubes wrapped with foil to prevent photolysis. $2cm^3$ of a suspension containing 200 second stage juveniles were added to give final concentrations of 0, 2, 4 and 8ppm of carbofuran. Each treatment was replicated three times. The test tubes were corked, placed in a rack and kept in a constant temperature room of $28^o \pm 1^oC$. After one and seven days exposure to carbofuran, the contents of each test tube were poured into a beaker containing distilled water (1 litre) and stirred. The nematodes were allowed to settle and the supernatant poured off. This was repeated three times to remove the pesticide. Aliquots containing about 100 juveniles were added into holes (1 inch deep) made around the roots of eggplant seedlings in 10cm plastic pots. The pots were arranged in the greenhouse in a fully randomized design. The temperature in the greenhouse ranged from $15^o - 37^oC$.

After fourteen days, plants were carefully removed from the pots; their roots were washed to remove adhering particles. The roots were stained and macerated with water ($100cm^3$) in a blender. The number of juveniles per root system were estimated from three $20cm^3$ aliquots taken from the $100cm^3$ suspension.

RESULTS

Posture and activity

Nematodes immersed in solutions of carbofuran and 3-hydroxycarbofuran were distorted (Figure 5.2.1). No tight coiling was observed. The effect of carbofuran on posture was greater than that of the metabolite as shown by the extent and the number of distorted nematodes (Table 5.1.3. and 5.1.2). The effect of 3-hydroxycarbofuran on posture of nematodes decreased with time (Table 5.1.3).

The activity of the nematodes was not affected by 3-hydroxycarbofuran as they appeared not to differ from the controls in terms of body movement (Table 5.1.1). Carbofuran influenced nematode activity which was related to concentration and time of exposure (Table 5.1.1). Hyperactivity was observed at all the concentrations of carbofuran used. At the highest concentration of 10ppm, the activity of the nematodes was reduced to below those of controls at 24 and 48 hours, but no reduction in activity was observed at the other concentrations used.

All the nematodes placed in distilled water for 6 hours resumed normal activity.

Effect of carbofuran on invasion of tomato roots by *M. incognita*

Carbofuran added at concentrations of 2μg - 8μg/g of sand significantly reduced root invasion ($p < 0.01$). (Table 5.1.4). Reduction in root invasion increased with increasing concentrations of carbofuran. The metabolite, 3-hydroxycarbofuran did not affect root invasion at any of the concentrations tested.

Invasion of second stage juveniles after exposure to different concentrations of carbofuran for different periods.

Exposure of juveniles to different concentrations of carbofuran

Table 5.1.1. Effects of carbofuran and its metabolite 3-hydroxycarbofuran on activity of second stage juveniles of *M. incognita*.

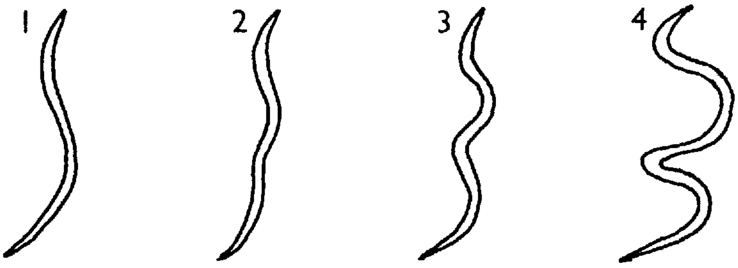
Treatments	Time after addition of pesticide (hrs) 2	4	6	24	48	6hr after removal from pesticide and immersed in water
Control (water)	5.0	5.0	5.0	5.0	5.0	5.0
Control 1% (v/v) acetone	5.0	5.0	5.0	5.0	5.0	5.0
Carbofuran 1ppm	5.0	5.0	6.0	5.6	5.6	5.0
2ppm	5.0	5.0	5.6	5.3	5.0	5.0
5ppm	5.0	5.3	5.3	5.0	5.0	5.0
10ppm	5.0	5.3	5.3	4.0	3.3	5.0
3 hydroxycarbofuran						
1ppm	5.0	5.0	5.0	5.0	5.0	5.0
2ppm	5.0	5.0	5.0	5.0	5.0	5.0
5ppm	5.0	5.0	5.0	5.0	5.0	5.0
10ppm	5.0	5.0	5.0	5.0	5.0	5.0

Table 5.1.2 Percentage of *M. incognita* juveniles which were distorted after exposure to different concentrations of carbofuran and 3-hydroxycarbofuran for varying times.

	Time after addition of pesticide (hrs)				
Treatments	2	4	6	24	48
Control (water)	0	0	0	0	0
Control 1% (v/v) acetone	0	0	0	0	0
Carbofuran 1ppm	0	50	80	100	100
2ppm	0	66	83	100	100
5ppm	0	73	90	100	100
10ppm	0	88	90	100	100
3 hydroxycarbofuran					
1ppm	0	0	0	0	0
2ppm	0	0	33	16	0
5ppm	0	0	53	46	23
10ppm	0	0	66	70	76

Figure 5.2.1

Ratings for distortion of shape of second stage juveniles of *M. incognita* after addition to pesticides for varying times.



1 - No distortion of shape observed

2 - Slightly distorted

3 - Moderately distorted

4 - Severely distorted

Table 5.1.3 Ratings for the distortion of second stage juveniles of *M. incognita* after addition to pesticides for varying times.

	Time after addition of pesticides (hrs)				
Treatments	2	4	6	24	48
Control (water)	1.0	1.0	1.0	1.0	1.0
Control 1% (v/v) acetone	1.0	1.0	1.0	1.0	1.0
Carbofuran 1ppm	1.0	3.0	4.0	4.0	4.0
2ppm	1.0	3.0	4.0	4.0	4.0
5ppm	1.0	4.0	4.0	4.0	4.0
10ppm	1.0	4.0	4.0	4.0	4.0
3 hydroxycarbofuran					
1ppm	1.0	1.0	1.0	1.0	1.0
2ppm	1.0	1.0	2.0	2.0	1.0
5ppm	1.0	1.0	3.0	2.0	2.0
10ppm	1.0	3.0	3.0	3.0	2.0

Table 5.1.4 Effect of soil application of carbofuran and 3-hydroxycarbofuran on invasion of tomato roots by second stage juveniles of *M. incognita*

Treatments	Mean number of juveniles in roots after 10 days 1) 2)
Control I 1% (w/v) phostrogen only	64.7
Control II 1% (v/v) acetone	61.2
Carbofuran 2μg/g of sand	19.0
4μg/g of sand	11.2
8μg/g of sand	5.0
3-hydroxycarbofuran	
2μg/g of sand	61.2
4μg/g of sand	61.5
8μg/g of sand	57.0
L.S.D. at 5%	9.2
L.S.D. at 1%	12.5

1) Means of 4 replicates

2) Initial population 350 nematodes.

Table 5.1.5 Number of nematodes in roots of eggplant after exposure to different concentrations of carbofuran for varying periods.

Number of nematodes in roots 1) 2)

Concentration in ppm					
Exposure time in days	0	2	4	8	
1	61.6	67.0	59.3	57.0	N.S.
7	53.3	42.0	45.3	41.3	N.S.

1) Means of 3 replicates.

2) Initial population inoculated 100 juveniles.

for varying times had no effect on their ability to invade roots after washing in water (Table 5.1.5). There was no significant difference between the number of nematodes in the control plants and those exposed to carbofuran ($p > 0.05$).

5.2 EFFECT OF CARBOFURAN ON THE MIGRATION OF SECOND STAGE JUVENILES OF *M. INCOGNITA*

The following experiment was set up to determine if the reduction in the invasion of roots by juveniles as observed in Sections 3.1 and by other workers (Di Sanzo (1969, 1973); Reddy and Seshadri (1971)), was due to the inability of the pesticide - treated juveniles to migrate towards the roots of host plants.

MATERIALS AND METHODS

Polyethylene tubes, 2cm long and 0.4cm in diameter, were covered at the end with a nylon mesh (45μ aperture) and half-filled with washed, air-dried sand (<180μ). The tubes were placed upright in small glass vials containing $1cm^3$ of carbofuran solution at concentrations of 1, 2, 4 and 8ppm; controls were placed in distilled water. Each treatment was replicated three times. Aliquots ($0.1cm^3$) of a suspension of freshly hatched second stage juveniles containing 50 juveniles were pipetted on to the surface of the sand. Each glass vial was covered with a plastic cork.

The glass vials were arranged on a tray and kept at $28^oC \pm 2^oC$ for 24 hours. Migration was then assessed by counting the number of juveniles that had passed through the sieve into the solution or water. This method is similar to that used by McLeod and Khair (1975).

RESULTS

Carbofuran at 1 - 2ppm had no significant effect on the ability of the juveniles to migrate through the sand column within 24 hours ($p > 0.05$) (Table 5.3.1). However, 4 and 8ppm significantly reduced the number which migrated through the column ($p \leq 0.01$).

Table 5.3.1 Effect of carbofuran on downward migration of second stage juveniles of *M. incognita* through a sand column.

Treatment	No. of juveniles migrating 1) through sand in 24 hrs.	% reduction or increase in migration cf control
Control (water)	34.3	-
Carbofuran 1ppm	36.0	+ 4.9
2ppm	31.6	- 7.8
4ppm	21.0	-38.7
8ppm	13.3	-61.2
L.S.D. at 5%	4.6	
L.S.D. at 1%	6.5	

1) Means of 3 replicates.

5.3 SYSTEMIC ACTION OF CARBOFURAN AGAINST SECOND STAGE JUVENILES OF *M. INCOGNITA* IN THE ROOTS OF EGGPLANT.

Carbofuran is known to possess systemic properties and applied to the soil it is taken up by roots and translocated to the aerial parts of the plant (Turner and Caro, 1973). The following experiment was designed to evaluate the systemic effect of carbofuran against *M. incognita* in the roots of eggplant.

MATERIALS AND METHODS.

Three-week old seedlings of eggplant were transplanted singly into 10cm plastic pots filled with heat-sterilized sand.

One week later each seedling was inoculated with 500 freshly hatched second stage juveniles of *M. incognita*. Three and seven days after inoculation plants were carefully removed from the pots and the roots washed thoroughly. These seedlings were transplanted singly into 22.5cm pots each filled with 5kg of sand to which carbofuran had been added at the rate of 4, 8 and 16kg ai, per ha (100, 200 and 400mg of Furadan 10G per pot). Control seedlings were transplanted into pots without carbofuran. Each treatment was replicated three times. Pots were arranged in a fully randomized design in the greenhouse (temperature range 16 - 39^{o}C.)

Seventeen days after transplanting into treated or untreated soil, the plants were harvested, their roots washed thoroughly, blotted dry and weighed. The roots were then stained, macerated and the number of females and juveniles per root system estimated as described previously.

RESULTS

Carbofuran applied to soil at different rates had no apparent effect on the development of females in the roots (Table 5.4.1).

Table 5.4.1. Effect of carbofuran on development of *M. incognita* in roots of eggplant

		% females 17 days after transplanting	
Treatment	Rate of carbofuran kg ai/ha	% females (3 days [1]) allowed for invasion)	% females (7 days allowed for invasion)
Plants from untreated soil into untreated	0	67.3	75.6
Plants from untreated soil into treated	4	62.6	74.2
"	8	64.8	70.0
"	16	69.2	71.6

1) Means of 3 replicates.

5.4 EFFECT OF CARBOFURAN ON HATCHING OF *M. INCOGNITA*

One of the ways by which carbofuran probably reduces the damaging effects of *M. incognita* to crop plants may be through inhibition of hatching. There is complete lack of information on the possible effects of carbofuran on egghatch of *M. incognita*.

The following experiment was therefore designed to evaluate the effects of carbofuran on hatching of *M. incognita* eggs.

MATERIALS AND METHODS

Egg masses of *M. incognita* of uniform size and colour were picked from tomato plants, six weeks after inoculation. Four egg masses containing an average of 350 eggs were placed in small sieves made from polyethylene rings, approximately 4mm thick, 2cm in diameter and covered at one end with a nylon cloth (45μ aperture). This small sieve was suspended in a syracuse dish containing $2cm^3$ of the nematicide solution (1, 2, 4 and 8ppm carbofuran); water served as the control. Each treatment was replicated three times. The experiment was done at $28^o \pm 1^oC$. The hatched juveniles were counted after 7 days; the remaining eggmasses were then washed in distilled water and the sieves resuspended in distilled water. After a further seven days, the number of hatched juveniles was again counted.

RESULTS

Hatching of *M. incognita* eggs were significantly ($p < 0.01$) reduced after exposure of eggmasses to 2, 4 and 8ppm of carbofuran for 7 days (Table 5.5.1). Following transfer to water the subsequent total hatch (in pesticide solution and in water) within the 14 day period was not significantly different between the treatments ($p > 0.05$).

Table 5.5.1 Effect of carbofuran on hatching of second stage juveniles of *M. incognita*

	Number of hatched juveniles.		
Treatment	No. of juveniles 1) in carbofuran (0-7 days)	No. of juveniles in water (7-14 days)	Total number of hatched juveniles
Control (water)	582.6	392.6	975.2
Carbofuran 1ppm	569.0	481.0	1050.0
2ppm	375.0	611.3	989.6
4ppm	314.3	771.0	1085.3
8ppm	164.6	828.6	993.2
L.S.D. at 5%	130.4		N.S.
L.S.D. at 1%	185.4		N.S.

1) Means of 3 replicates

5.5 EFFECT OF BARE ROOT DIPPING OF EGGPLANT IN CARBOFURAN ON INVASION BY *M. INCOGNITA*

The possible role of carbofuran present in the roots of plants in treated soil in preventing root invasion by *M. incognita* has not been evaluated. This experiment therefore is intended to assess the importance of carbofuran within the roots of eggplants in preventing root invasion by *M. incognita*.

MATERIALS AND METHODS

Twenty four three-week old seedlings of eggplant of uniform size were uprooted and their roots washed thoroughly. These seedlings were grouped into four batches of six seedlings and each batch was dipped in the different concentrations of carbofuran (2, 4 and 8ppm) and in control (water). Twenty-four hours later the roots were washed thoroughly and the seedlings transplanted singly into 22.5cm plastic pots filled with sterilized soil. Pots were arranged in a fully randomized design in the greenhouse (temperature ranged between 17^{o} - $40^{o}C$ during the experiment). At 2 and 9 days after transplanting, 600 freshly hatched second stage juveniles of *M. incognita* were added to the pots of three plants from each batch dipped in the same concentration of carbofuran or in control. Two weeks after inoculation, the plants were removed from the pots and the roots washed free of adhering soil. The roots were dried on blotting paper and weighed. Whole root systems were stained in 0.1% (w/v) cotton-blue in lactophenol and macerated in a blender. Estimates of the number of juveniles in the roots were determined from aliquots from the suspension.

RESULTS

There was no significant difference between the total number of nematodes in the roots of plants dipped in different concentrations of carbofuran and the control ($p > 0.05$), for plants inoculated at 2 or 9 days after replanting (Table 5.6.1). However, in plants inoculated after two days, the percentage of nematodes which had developed to the saccate stage was less in the roots dipped in carbofuran.

Table 5.6.1. <u>Effect of bare root dip in carbofuran solution on root invasion by second stage juveniles of *M. incognita*.</u>

Treatments	No. of nematodes in roots 14 days after replanting			
	Plants inoculated 2 days after replanting.		Plants inoculated 9 days after replanting	
	Total nematodes [1]	% saccate nematodes	Total nematodes	% saccate nematodes
Control (water)	366.1	64	324	57
Carbofuran at 2ppm	358.6	47	339.6	59
4ppm	274.0	42	294.3	53
8ppm	304.3	36	281.0	56
L.S.D. at 5%	N.S.		N.S.	
L.S.D. at 1%	N.S.		N.S.	

1) Means of 3 replicates

5.6 BEHAVIOURAL RESPONSES OF SECOND STAGE JUVENILES OF *M. INCOGNITA* TO CARBOFURAN ON AGAR PLATES.

The behaviour of second stage juveniles of *M. incognita* on agar plates containing carbofuran has been studied by Di Sanzo (1969, 1973). He observed that nematodes placed about 4cm away from the roots of germinating tomato seedlings did not invade or feed because they were disorientated. Nematodes were motile but moved randomly. When the nematodes were deposited directly on tomato roots most of them migrated away and did not invade. This suggests that the impairment of the sensory organs may be involved in carbofuran action. However, the above study was carried out using the high concentration of 10ppm carbofuran. In the present work studies with the lower concentrations of carbofuran were done as these were likely to give a better indication of the mechanism by which carbofuran controls *M. incognita* at rates used under field conditions.

MATERIALS AND METHODS.

Tomato seeds (var Moneymaker) were surface sterilized in 3% (v/v) sodium hypochlorite for 20 minutes and washed four times in sterile water. The seeds were then placed in petri dishes lined with moist filter papers;five days later germinating seeds were transferred to petri dishes (9cm diameter) containing 1% (w/v) Ion agar to which carbofuran had been added just prior to solidification to give concentrations of 1, 2, 4 and 8ppm. Petri dishes containing agar without carbofuran served as control. Each treatment was replicated three times.

A tomato seedling was placed 1cm from the perimeter of each dish. Fifty juveniles in 0.5ml of water were added to each plate around the roots of the seedlings. The plates were kept at $28^{o}C \pm 1^{o}C$.

Table 5.7.1. Effect of carbofuran on movement and invasion of second stage juveniles of *M. incognita* on agar plates with tomato seedlings.

	No. of nematodes away from seedlings 1) 2)				Nematodes in roots after 7 days	
Treatments	Type of movement	6hrs	24hrs	48hrs	Total number in roots	% nematodes in roots
Control (Agar only)	Directional	0	0	0	29	58
Carbofuran 1ppm	non-directional	12	23	38	12	24
2ppm	non-directional	8	15	21	8	16
4ppm	non-directional	3	14	16	0	0
8ppm	non-directional	0	6	12	0	0

1) Means of 3 replicates

2) Initial number inoculated (50 juveniles)

Observations on the nematode behaviour namely movement and position in relation to the roots, were made after 6, 24 and 48 hours. On the 7th day the roots were stained and the number of nematodes counted as described previously. During the experiment, $2cm^3$ of 0.1% (w/v) phostrogen solution was added on the 1st and 4th day.

RESULTS

Nematodes placed on agar plates without carbofuran remained near the roots and were observed attempting to feed shortly after transfer (Table 5.7.1). Fifty-eight percent of the nematodes invaded within the 7-day period. Nematodes placed on plates containing carbofuran did not remain near the roots and after six hours, many of the nematodes on agar plates containing carbofuran at 1 and 2ppm had moved away (Table 5.7.1). Movement was seen to be non-directional. At higher concentrations of carbofuran, there was less movement away from roots and most of the nematodes in plates containing 8ppm of carbofuran remained near the roots. They were mobile but were not seen to feed and appeared disorientated. No nematodes were found in the roots of seedlings in plates containing 4 and 8ppm of carbofuran (Table 5.7.1).

DISCUSSION

The effects of carbofuran and its metabolite on the posture and activity of *M. incognita* juveniles shown in the above experiments are in agreement with the view that inhibition of acetylcholinesterase is the mode of action of organophosphate and carbamate nematicides (Corbett, 1974; Croll, 1975; Le Patourel and Wright, 1976) resulting in the impairment of neuromuscular coordination. Several other workers have also observed that the posture of nematodes are affected by non-volatile nematicides (Keetch, 1974; Le Patourel and Wright, 1974a, 1974b; Bunt, 1975; Batterby *et al*, 1977).

The results indicated that carbofuran was more toxic than the metabolite (Tables 5.1.2 and 5.1.3). The decrease in the number of distorted nematodes with time and the extent to which the nematodes were distorted (Tables 5.1.2 and 5.1.3) might have been due to the breakdown of 3-hydroxycarbofuran under the experimental conditions. The inability of 3-hydroxycarbofuran to prevent root invasion at the different concentrations used (Table 5.1.4) suggests that it plays no role in the control of *M. incognita* at rates of carbofuran used under field conditions. The formation of 3-hydroxycarbofuran therefore represents a detoxification process in carbofuran degradation. The high activity shown by the nematodes in carbofuran even at 5ppm is in contrast with the observed effects of aldicarb or oxamyl on body movement of *M. incognita* (Nelmes and Keereewan, 1970; Wright *et al*, 1980). This suggests that carbofuran is probably less toxic to *M. incognita* than aldicarb or oxamyl.

Carbofuran significantly reduced migration of 2nd stage juveniles at 4 and 8ppm which is much higher than the effective concentration in soil water that can be found under field conditions, and this impairment of nematode migration may be of no significance in the control of *M. incognita* at recommended rates of carbofuran.

Similarly, the high infectivity of nematodes after exposure to different concentrations of carbofuran for varying times indicates that the nematodes could rapidly recover from the effects of the nematicide and still have sufficient energy for invasion, despite the fact that they were mobile most of the time at the lower pesticide concentrations. This is possibly because root invasion is not thought to consume much energy (Van Gundy *et al.*, 1967).

Fewer saccate nematodes were found in the carbofuran-dipped roots inoculated 2 days after replanting which may be due to a temporary delay in invasion because of the presence of the nematicide in the root. This is supported by the observation that carbofuran had no effect on the development of nematodes already in the roots of egg-plant (Table 5.4.1) even when transplanted into soil treated with carbofuran at 16kg ai/ha . These results indicate that carbofuran acts principally against *M. incognita* in the soil.

The results obtained on systemic action and that of bare root dipping tests indicate that carbofuran persists for a very short time in the roots probably partly because it is rapidly translocated to the leaves and also because it is rapidly converted into non toxic metabolites. Similarly, Hague and Pain (1973) could not find any evidence that al-dicarb, thionazin and methomyl affected *Globodera rostochiensis* in the roots of potato. Several other studies have demonstrated that once in the plant, nematodes are less sensitive to pesticides (White-head, 1973; Bromilow and Lord, 1979). The control of

M. incognita when carbofuran was applied as post plant treatments (Sivakumar *et al.*, 1974, 1976) is therefore due to its effects on nematodes in the soil rather than in the plant.

The reduction in egg hatch is probably one of the other ways in which carbofuran helps to reduce the damaging effects of *M. incognita* to crop plants as the emergence of infective juveniles is delayed. However, the total hatch obtained was similar for all the treatments indicating that the eggs were not irreversibly affected by carbofuran. Similar effects have been found for oxamyl on egg hatching of *M. incognita* (Wright *et al.*, 1980), and Hague and Pain (1973) found that the total hatch of *Globodera rostochiensis* was not decreased by aldicarb, thionazin and methomyl. But McLeod and Khair (1975) reported that when eggmasses of *M. incognita* were transferred from nematicides which suppressed hatch to water, hatching occurred, but aldicarb, phenamiphos, ethroprophos and thionazin significantly reduced total hatch. Carbofuran has been reported to affect hatching and emergence of *Heterodera schachtii* (Steele, 1977). The effect was also reversible.

The results obtained on the behaviour of *M. incognita* juveniles on agar plates are broadly similar with that reported by Di Sanzo (1969, 1973) for *P. penetrans* and *M. incognita*. The results, however, reflect its possible effects on nematodes at lower concentrations in the soil.

The migration of nematodes away from the roots on agar plates containing carbofuran particularly at the lower concentrations indicated that they were probably unable to detect the presence of roots. This suggests the possible impairment of sensory organs. Acetylcholinesterase has been detected histochemically in the amphids, phasmids and cephalic papillae of *Dipetalonema vitae* (McLaren, 1972) and *Caenorhabditis elegans*

(Pertel *et al.*, 1972) and the possible disruption of such organs by the effect of carbofuran on acetylcholine esterase contained in them could result in the observed behaviour of the nematodes. A similar suggestion has been made for effects on nematode behaviour of *M. incognita* by oxamyl (Wright *et al.*, 1980). Ward (1973) has shown that sensory receptors in the head are involved in the orientation behaviour of *C. elegans*. However, the involvement of poor neuromuscular coordination may also be involved as this might affect the behavioural sequences necessary for invasion (Lee and Atkinson, 1976) and egg hatching (Wallace, 1968). The results of this study suggest that this probably operates more at the high concentrations of carbofuran.

SECTION 6

6.0 FACTORS INFLUENCING THE EFFECTS OF FERTILIZERS ON *M. INCOGNITA* INFECTING EGGPLANT.

INTRODUCTION

The results obtained by Oteifa (1952) on the influence of potassium in relation to infection of lima beans by *M. incognita* suggest that the level of application of certain fertilizers and the population density of the nematodes are the important factors influencing damage to the crop plant. The damaging effects on lima beans being reduced when potassium fertilizer was applied in large amounts to soil which was moderately infested by the nematodes whereas the application of high levels of potassium fertilizers had no effect on damage caused by *M. incognita* on the crop at high soil infestation levels.

As seen from the results in Section 3.3, the growth of lettuce and carrots infected with *M. incognita* was enhanced by the application of fertilizers but the actual crop loss due to the nematodes was unaltered (Tables 3.4.1 and 3.4.2). It was not clear if this was due to the crop type or plant age at the time of infection. A study was therefore needed to determine the importance of plant age in the relationship between fertilizer application and the damaging effects of *M. incognita* to crop plants.

The experiments reported in this section were designed to determine the influence of varying the levels and proportion of NPK fertilizers; high soil population density of the nematodes and the plant age on the effects of fertilizers on *M. incognita* on eggplant.

6.1 INFLUENCE OF VARYING RATES OF FERTILIZERS

MATERIALS AND METHODS

Forty 22.5cm plastic pots were each filled with 5kg of heat sterilized soil. NPK was added to the pots at transplanting at the rates of 0, 750kg/ha (18.29g/pot) and 1500kg/ha (36.58g/pot). A treatment in which fertilizers was applied at 1500kg/ha in two equal doses at transplanting and three weeks later, was also included. A treatment involving the combined application of carbofuran at 8kg ai/ha and fertilizers at 750kg/ha (as used in Section 3) was included for comparison.

Four-week old seedlings of eggplant were transplanted singly into each pot and half of these were inoculated with 3,000 freshly hatched second stage juveniles of *M. incognita*. Each treatment was replicated four times. Pots were arranged in a fully randomized design in the greenhouse (temp range (16^o-39^oC). Six weeks after inoculation additional fertilizers were given to pots with fertilizer at the rate of 9.14g/pot. The experiment was terminated sixty-five days after inoculation. The fresh weight of tops and roots were taken and the number of developing fruits counted. Root systems were rated for the incidence of root galling and the percentage of roots galled. The soil population of second stage juveniles was also estimated.

RESULTS

Based on the fresh weight of tops, the growth of infected and uninfected plants growing in soils given the various fertilizer treatments were all significantly ($p < 0.01$) increased over those of plants

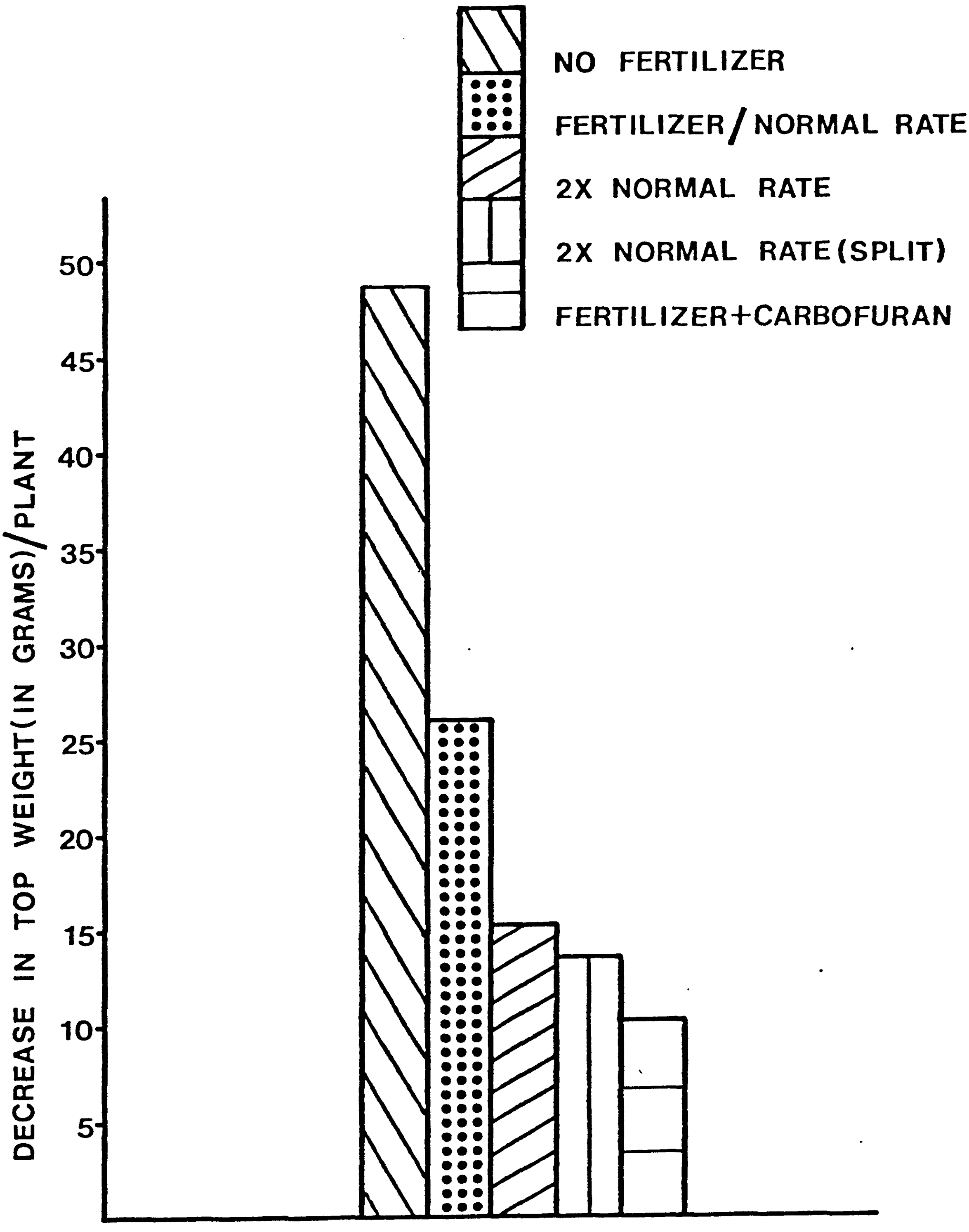


Fig. 6.2.1. Reduction in top growth of eggplant infected with <u>M.incognita</u> as influenced by varying fertilizer rates. (Differences between the mean top weights of uninfected and infected plants).

Plate 2: Effects of varying rates of fertilizers on the growth of eggplant infected with M. incognita. (40 days after inoculation).

A - Fertilizers (2 x normal rate) + Nematodes

B - Fertilizers (2 x normal rate) - Nematodes

C - Fertilizers (2 x normal rate, split) + Nematodes

D - Fertilizers (2 x normal rate, split) - Nematodes

E - Fertilizers + Nematicide + Nematodes

F - Fertilizers + Nematicide - Nematodes

G - Fertilizers (at normal rate) + Nematodes

H - Fertilizers (at normal rate) - Nematodes

I - No fertilizers + Nematodes

J - No fertilizers - Nematodes


A
B
C
D
E
F
G
H
I
J

in soil not given fertilizers (Table 6.2.1). Amongst the infected plants, the greatest top weight was obtained for plants in soil treated with carbofuran (Table 6.2.1) but this was not different from those of plants given an extra dose of fertilizers ($p > 0.05$). The fresh weight of tops of infected plants were all significantly($p < 0.05$) reduced by nematode infection, except for infected plants in carbofuran treated soil (Table 6.2.1.). The differences in fresh weight of tops between uninfected and infected plants in soil given the different treatments are summarised in Figure 6.2.1. The number of fruits on infected plants given twice the rate of fertilizers and those in soil treated with carbofuran were significantly greater than those on plants in soil given the normal rate of fertilizers ($p < 0.05$) (Table 6.2.1.). The greatest number of fruits were found on plants in soil given fertilizers in split doses and those in soil treated with carbofuran and were significantly ($p < 0.05$) greater than the number on plants given twice the rate of fertilizers at transplanting. No fruits were found on infected plants in soil without fertilizers.

The fresh root weights of infected plants were significantly ($p < 0.01$) greater than those of uninfected plants given the same treatment, except for plants in carbofuran-treated soil where there was no significant difference, ($p > 0.05$).

Nearly all roots of infected plants in soil without fertilizers were severely galled, but in soils given fertilizers at twice the normal rate (1500kg/ha) and in carbofuran treated soil, up to 30% of the roots were free from galls, (Table 6.2.1.).

The soil populations of juveniles obtained for the various treatments were significantly ($p < 0.01$) greater in soils given an extra dose of fertilizer (Table 6.2.1).

Table 6.2.1. Influence of varying rates of NPK fertilizers on the growth of eggplant infected with *M. incognita*.

Treatments	Fresh weight of tops (g) 1)	% reduction in growth	Average number of fruits/ plant	Fresh weight of roots (g)	Galling index	2) Scores for % of roots galled	No. of juvenile /200g of soil.
Fertilizers (2N) - Nematodes	85.4	15.1	2.8	16.6	5.0	3.0	2050
Fertilizers (2N) - Nematodes	100.6		4.2	48.6	0	0	0
Fertilizers (2N)(Split) + Nematodes	88.4	13.2	4.0	50.6	5.0	3.0	2400
Fertilizers (2N)(Split) - Nematodes	101.8		4.0	40.2	0	0	0
Fertilizers (N) + Nematodes	69.4	27.4	1.5	42.5	5.0	4.0	1813
Fertilizers (N) - Nematodes	95.6		3.0	33.2	0		0
Carbofuran + Fertilizers + Nematodes	92.6	10.3	4.2	36.3	4.0	3.0	1650
Carbofuran + Fertilizers - Nematodes	103.2		4.7	33.4	0	0	0
None + Nematodes	9.6	83.5	0.0	8.0	6.0	5.0	540
None - Nematodes	58.4		0.5	13.9			0
L.S.D. at 5%	12.1		1.2	5.2			596.6
L.S.D. at 1%	16.3		1.6	7.0			825.1

1) Means of replicates

2) See Section 4.2.

6.2 INFLUENCE OF HIGH SOIL POPULATION DENSITY

The inoculum levels of *M. incognita* used in the experiments reported in previous sections can be regarded as low to moderate. In order to better assess the potential of fertilizer application as a means of minimizing losses caused by *M. incognita* on vegetables, their effectiveness should also be tested at high levels of infestation.

In the following experiment, the influence of fertilizers on the growth of eggplant in soil heavily infested with *M. incognita* was studied.

MATERIALS AND METHODS

Infested soil which was not treated with carbofuran taken from pots used in the previous experiment (Section 6.1) was mixed with appropriate amounts of heat sterilized soil to give a population density of 600 second stage juveniles per 200g of soil. Plastic pots 22.5cm in diameter, were each filled with 5kg of the infested soil that is 15,000 second stage juveniles per pot. Five kilograms of heat sterilized soil was added to an equal number of pots. NPK fertilizer was added to these pots at 0, 750kg/ha (18.29g/pot) and 1500kg/ha (36.58/pot).

Four-week old seedlings of eggplant were then transplanted singly into each pot. There were four replicates per treatment. The pots were arranged in a fully randomized design in the greenhouse (temperature range (15 - 40^{o}C)). Six weeks after inoculation, 9.14g of fertilizers was added to each of the pots given fertilizers at transplanting. Pots were watered whenever necessary.

Sixty-five days after inoculation, the fresh weight of tops and

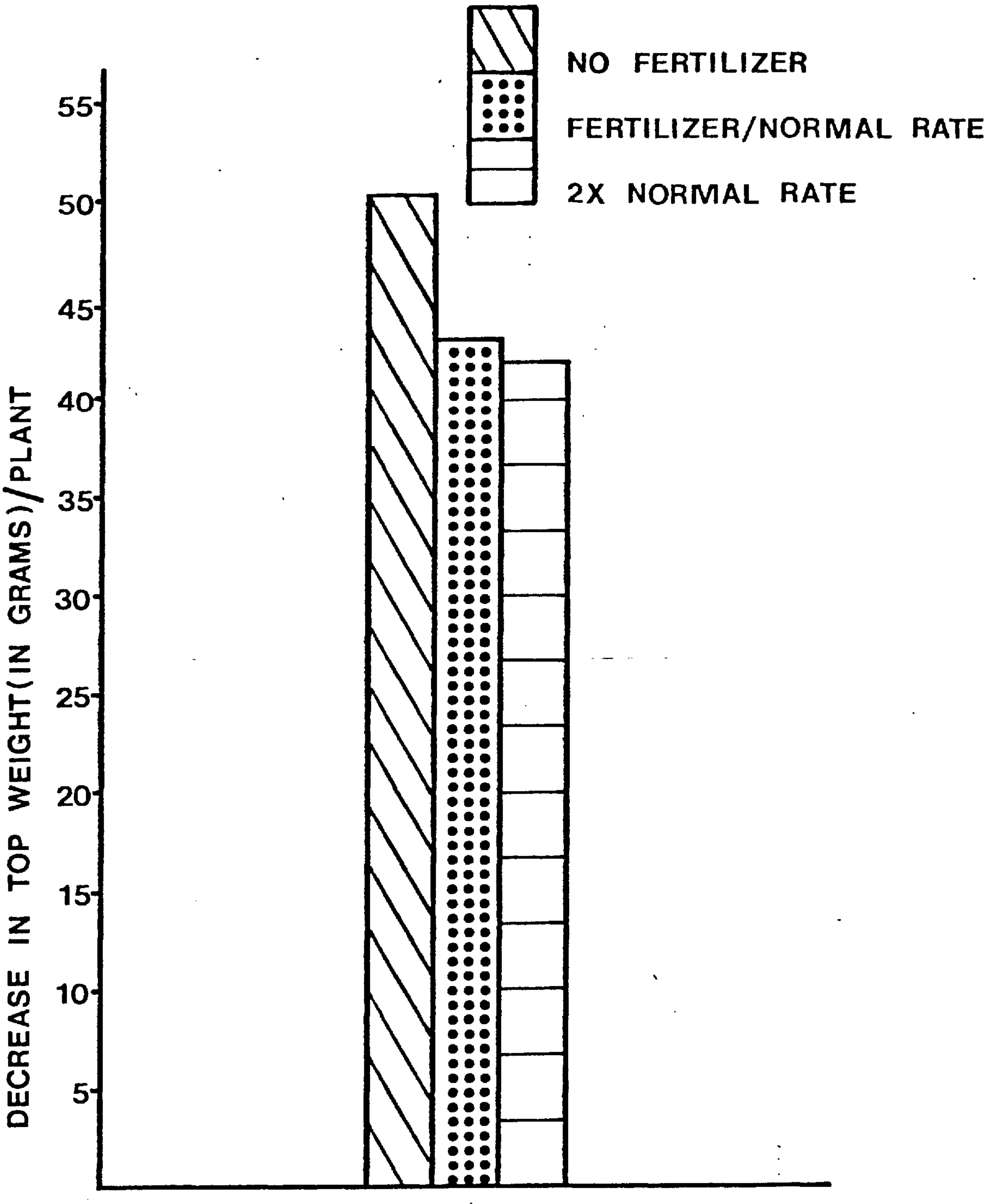


Fig. 6.3.1. Reduction in top growth of eggplant infected with M. incognita at high soil population density.

(Differences between the mean top weights of uninfected and infected plants).

roots was recorded. The numbers of developing fruits per plant were counted prior to harvesting. The incidence of root galling and the proportion of roots galled were also assessed, and the soil population of second stage juveniles estimated.

RESULTS

After four weeks from transplanting all the seedlings in infested soil which had not been given fertilizers had died. Plant growth based on fresh weight of tops was significantly ($p < 0.01$) increased by the application of fertilizers with the greatest top weight being obtained for plants in soil given an extra dose of fertilizers (Table 6.3.1). The top weight of infected plants was significantly ($p \leq 0.01$) reduced by nematode infection, irrespective of treatment. The mean difference in fresh weight of tops between uninfected and infected plants were 42.0, 43.1 and 50.4 grams, respectively, for plants grown in soil given an extra dose of fertilizers, the normal rate of fertilizer or no fertilizer.

At the termination of the experiment, no fruits were found on infected plants, but plants growing in noninfested soil had developing fruits (Table 6.3.1). All the roots of infected plants were severely galled with less than 10% of the roots free from galls. (Table 6.3.1). There was no significant difference between the incidence of root galling at the two rates of fertilizer application ($p > 0.05$).

The fresh weight of roots of infected plants were significantly ($p < 0.05$) greater than those of uninfected plants in soil given twice the normal rate of fertilizer but no significant difference was found between the root weights of infected and uninfected plants in soil given fertilizers at the normal rate ($p > 0.05$).

Table 6.3.1. Influence of fertilizers on the growth of eggplant in soil heavily infested with *M. incognita*

Treatment	Fresh weight of tops in grams	% reduction in growth	Average number of fruits per plant	Fresh weight of roots	Galling Index	No. of juveniles L2/200g of soil
Fertilizer (2N) + Nematodes	65.3	39.3	0	46.4	6.0	3500
Fertilizer (2N) - Nematodes	107.3		4.0	38.4	0	0
Fertilizer (N) + Nematodes	48.5	46.9	0	33.6	6.0	3098
Fertilizer (N) - Nematodes	91.6		3.0	31.2	0	0
None + Nematodes	0	100	0	0	-	-
None - Nematodes	50.4		0	14.6	0	0
L.S.D. 5%	11.4			5.39		N.S.
L.S.D. 1%	15.8			7.45		

There was no significant difference between the number of second stage juveniles extracted from pots given fertilizers at the two rates ($p > 0.05$).

6.3 INFLUENCE OF PLANT AGE.

The following experiment was designed to compare the influence of fertilizers on the damaging effects of *M. incognita* to one week and four week old seedlings of eggplant.

MATERIALS AND METHODS.

Thirty-two 22.5cm plastic pots were each filled with 5kg of heat sterilized soil; these pots were divided into two batches of 16 pots each. Half of the pots in each batch were given fertilizers at 750kg/ha (18.29g/pot); the rest were untreated. Five seeds of eggplant were sown in each of the sixteen pots of one batch. One week after germination, the seedlings were thinned to one per pot. Eight of these pots were inoculated with 500 second stage juveniles each. The remaining eight pots were not inoculated with nematodes. Four week old seedlings were transplanted singly into the remaining sixteen pots (8 treated and 8 untreated), and were inoculated in the manner described above. Details of the various treatments are given in Table 6.4.2.

Another set of twelve seedlings (6 one-week old and 6 four week old) which were inoculated as described above were included to assess the incidence of root galling three weeks after inoculation. Half of the seedlings in each age group were given fertilizers at 750kg/ha (18.29g/pot) while the others were not treated. Pots were arranged in the greenhouse in a fully randomized design. (Temperature ranged from 16-38^{o}C).

Three weeks after inoculation, the twelve additional plants were harvested and the number of galls per root system and per gram of root were assessed. Six weeks after inoculation additional fertilizers were added to pots given fertilizers initially at the rate of 9.14g per pot.

Table 6.4.1. Influence of fertilizers on root galling of eggplant infected with *M. incognita* at one and four weeks after germination.

Data taken 3 weeks after inoculation.

Treatments	One week old seedlings		Four week old seedlings	
	No. of galls per root system 1)	No. of galls/gram of root	No. of galls per root system	No. of galls per gram of root
Fertilizers + Nematodes	91.3	30.3	299.0	25.8
None + Nematodes	42.3	42.6	278.0	43.6
L.S.D. 0.05	37.5	N.S.	N.S.	8.5
L.S.D. 0.01	62.5	N.S.	N.S.	14.2

1) Means of 3 replicates

Table 6.4.2. Influence of fertilizers on the growth of eggplant infected with *M. incognita* at one and four weeks after germination.

Data taken at harvest

Treatments	One week old seedlings			Four week old seedlings		
	Fresh weight of tops 1)	Fresh weight of roots	Galling Index	Fresh weight of tops	Fresh weight of roots	Galling Index
Fertilizer + Nematodes	21.70	10.10	5.0	89.60	43.60	5.0
Fertilizer - Nematodes	37.80	9.35	0.0	84.67	34.30	0.0
None + Nematodes	8.70	3.70	5.0	32.60	15.38	5.0
None - Nematodes	25.85	5.20	0.0	51.70	13.90	0.0
L.S.D. at 0.05	6.50	1.83	N.S.	11.45	4.85	N.S.
L.S.D. at 0.01	9.10	2.13	N.S.	16.06	6.71	N.S.

1) Means of 4 replicates

The experiment was terminated sixty five days after inoculation and the fresh weight of the tops and roots recorded. The incidence of root galling was rated using a modification of the method described by Daulton and Nausbaum (1961).

RESULTS.

Plants inoculated one week after germination.

At the termination of the experiment, the fresh top weight of plants inoculated one week after germination, and given fertilizers was reduced by 42.6%, whereas for plants in soil without fertilizers, a reduction in top growth of 69.8% was found (Table 6.4.2,). Fertilizer X nematode interaction for top growth was not significant indicating that the effect of the nematodes on plant growth was not reduced by fertilizer application .

At three weeks after inoculation, there was no significant ($p > 0.05$) difference between the number of galls per root system for plants in fertilized and unfertilized soil (Table 6.4.1). Similarly at the termination of the experiment, the gall ratings was the same for plants in soil given fertilizers and unfertilized soil ($p > 0.05$) (Table 6.4.2).

Plants inoculated four weeks after germination.

For plants inoculated four weeks after germination, the top weight of infected plants in soil given fertilizers was not reduced by nematode infection ($p > 0.05$), but in contrast the fresh top weight of infected plants in unfertilized soil was reduced by 36.9% (Table 6.4.2). The interaction, fertilizer X nematode was significant, indicating that damage to the plant was markedly reduced by the application of fertilizers.

At three weeks after inoculation, the number of galls per gram of root was significantly ($p < 0.01$) less on the roots of plants in soil given fertilizers (Table 6.4.1). At the termination of the experiment, there was no significant difference between the incidence of root galling for plants in fertilized and unfertilized soil.

6.4 INFLUENCE OF VARIOUS COMBINATIONS OF N, P & K FERTILIZERS ON THE EFFECTS OF *M. INCOGNITA* ON EGGPLANT.

The following experiment was designed to examine the relationship between various combinations of N, P and K fertilizers and damage caused by *M. incognita* on eggplant.

MATERIALS AND METHODS.

Fifty-six 22.5cm plastic pots were each filled with 5kg of heat sterilized washed sand. All pots were given an initial dose of $10cm^3$ of 10% (w/v) phostrogen solution to provide micronutrients for the plants. Various combinations of N, P and K fertilizer as shown in Table 6.5.1 were added to the pots in four replications. The sources of nitrogen, phosphorus and potassium were $(NH_4)_2SO_4$, superphosphate and potassium sulphate respectively.

Four week old seedlings of eggplant were transplanted singly into each pot and half of the pots were inoculated with 1000 freshly hatched second stage juveniles per pot. The remaining pots were not inoculated. Two additional plants per treatment for the various combinations of N, P, and K were included to assess incidence of root galling at 24 days after inoculation. All the pots were arranged in a fully randomized design in the greenhouse (temperature range 16 - 39^{o}C). Twenty four days after inoculation, the additional plants included were harvested and the number of galls per root system counted. Sixty-five days after inoculation, the remaining plants were harvested and the fresh top and root weight, and the incidence of root galling recorded as described previously.

RESULTS

The poorest growth, based on fresh top weight, was obtained when

Table 6.5.1. Influence of various combinations of N, P and K fertilizers on the growth of eggplant infected with *M. incognita*.

Treatments	Fresh weight of tops 1)	% reduction in growth	Fresh weight of roots	Galling index	*2) No. of galls/ root system	*No of galls 1 g of root
$N\text{-}P_1K_1$ + Nematodes	12.20	63.25	6.20	5.0	284.5	78.3
$N\text{-}P_1K_1$ - Nematodes	33.20		5.40	0.0	0.0	0.0
$N_1P\text{-}K_1$ + Nematodes	17.70	56.97	8.90	5.0	302.5	65.9
$N_1P\text{-}K_1$ - Nematodes	36.10		7.15	0.0	0.0	0.0
$N_1P_1K\text{-}$ + Nematodes	28.30	35.97	8.40	5.0	306.0	52.5
$N_1P_1K\text{-}$ - Nematodes	44.20		7.20	0.0	0.0	0.0
$N_1P_1K_1$ + Nematodes	46.10	23.76	12.30	5.0	292.0	48.65
$N_1P_1K_1$ - Nematodes	52.60		10.20	0.0	0.0	0.0
$N_2P_1K_1$ + Nematodes	41.85	19.26	11.85	5.0	340.5	38.9
$N_2P_1K_1$ - Nematodes	51.80		10.20	0.0	0.0	0.0
$N_1P_2K_1$ + Nematodes	39.50	25.8	13.30	5.0	457.0	57.7
$N_1P_2K_1$ - Nematodes	53.40		11.20	0.0	0.0	0.0
$N_1P_1K_2$ + Nematodes	39.80	20.71	12.90	5.0	359.0	40.0
$N_1P_1K_2$ - Nematodes	50.20		11.90	0.0	0.0	0.0
L.S.D. at 5%	7.25		3.00	N.S.		
L.S.D. at 1%	9.76		4.05	N.S.		

1) Mean of 4 replicates

2) Mean of 2 replicates

N_1 - $(NH_4)_2SO_4$ applied at the rate of 250kg/ha. (6.09g/pot)

P_1 - Superphosphate (P_2O_5) applied at the rate of 250kg/ha. (6.09g per pot)

K_1 - Potassium sulphate (K_2SO_4) applied at the rate of 125kg/ha. (3.04g/pot).

infected plants were grown in sand deficient in nitrogen (Table 6.5.1). The fresh top weight of infected plants in sand deficient in either N, P or K was significantly ($p < 0.01$) less than those of plants given all the nutrients ($N_1P_1K_1$). Similarly, the fresh top weight of uninfected plants in sand deficient in N, P or K was significantly less ($p < 0.05$) than those of plants given the $N_1P_1K_1$ treatment.

The growth of plants was significantly ($p < 0.05$) reduced by nematode infection, irrespective of treatment. Based on the mean difference between the top weights of uninfected and infected plants, the greatest reduction was obtained for plants in sand deficient in nitrogen (21.0 grams) while the least reduction occurred when plants were given an extra dose of nitrogen (9.95 grams). The root weights of infected plants were generally greater than those of uninfected plants. There was no significant difference between the root weights of infected plants deficient in N, P or K ($p > 0.05$). The addition of complete nutrients to sand ($N_1P_1K_1$) and increasing the dose of N P and K resulted in significant ($p < 0.05$) increases in root weights of infected plants over those of plants in sand deficient in nutrients. (Table 6.5.1).

No significant difference was found between the incidence of root galling for plants given the various combinations of fertilizers ($p > 0.05$).

DISCUSSION

The results (Table 6.2.1) have shown that the action of fertilizers against the damaging effects of *M. incognita* can be enhanced by increasing the level of fertilizer application. The results obtained from the experiment reported in Section 4.1 indicated that the application of fertilizers at the highest rate might have some toxic effect on the nematodes. However, since a similar reduction in plant growth was obtained when the same amount of fertilizer was applied in two doses, the effect may not be due to the toxic effect of the fertilizers on the nematodes. The application of an extra dose may have further enhanced the capacity of the plants to compensate for damaged roots. The larger root system may have also ensured that more roots were able to escape nematode infection at the later stages of the plant's growth. The greater percentage of roots not galled for plants in soil given an extra dose of fertilizers when compared to that found for plants in soil given the lower dose of fertilizers (Table 6.2.1) seems to suggest this. It is however important to note that the growth of infected plants was not completely restored as the plant's growth was significantly reduced by nematode infection, when plants were given an extra dose of fertilizer (Table 6.2.1).

In soils heavily infected with *M. incognita*, the ineffectiveness of fertilizers against the damaging effects of *M. incognita* shows that newly formed roots may have been attacked soon after they were formed. These plants therefore lacked the capacity to utilize available nutrients in the soil despite enhanced compensatory root growth made possible by the application of fertilizers. This result is similar to that obtained by Oteifa (1952) who found that the damaging effects of *M. incognita* on lima beans was reduced only when high rates of potassium fertilizers were applied to soil that was moderately infested.

Plant age as a factor influencing the effects of *M. incognita* on crop plants has been shown from the results obtained (Table 6.4.1 and 6.4.2). Plants infected one week after germination, lacked the capacity to sufficiently compensate rapidly for root damage as shown by the similar reduction in plant growth in soil given fertilizers and in unfertilized soil. The similar number of galls per gram of root for plants grown in both soils at 24 days after inoculation confirms this (Table 6.4.1). In contrast the growth of eggplant inoculated at four weeks after germination was not reduced at this level of infection. The growth of these plants may have been stimulated in response to the attack by low population of *M. incognita*. Wallace, (1971b) found that the growth of tomato was stimulated at low populations of *M. javanica*. He concluded that the opposing influences of root destruction and metabolic sink formation on one hand and root generation and growth hormone production on the other are imagined to work simultaneously. If the inhibitory process predominate, yield or growth of the plant is reduced, but there is less or no growth reduction if the stimulatory components predominate. There may in fact be an increase in growth of infected plants if growth substances are produced in excess. But Madamba *et al.* (1965) concluded that the stimulatory effect of *Meloidogyne* spp. on crop plants was due to increased root growth which gave added powers of water and mineral uptake.

Based on the above, it could be concluded from the results obtained from the experiment on the influence of plant age (Table 6.4.1), that the young seedlings probably lacked enough metabolites to compensate sufficiently for damaged roots or lacked the hormones necessary for stimulatory growth as suggested by Wallace, (1971b).

The greater reduction in plant growth in the absence of nitrogen indicates that it is very important in compensatory root growth. The number of galls per root system obtained for plants given the various treatments (Table 6.5.1) indicates that toxicity of the fertilizers to the nematodes was not involved. Shafiee and Jenkins (1962) also found that the top weight of pepper infected with *P. penetrans* was most markedly reduced in plants deficient in nitrogen. This may be related to the important role of nitrogen in chlorophyll production which is essential for photosynthesis, the process whereby photosynthates necessary for root formation are formed. Although increasing the amounts of nitrogen and potassium further lessened the damaging effects of *M. incognita* on the plants, the importance of maintaining a proper nutritional balance in soil infested with root-knot nematodes is indicated. The greater reduction in plant growth in sand deficient in any of the nutrients seem to emphasize this.

SECTION 7

FACTORS INFLUENCING THE EFFICACY OF CARBOFURAN AGAINST *M. INCOGNITA*

INTRODUCTION

Carbofuran like other non-fumigant nematicides, is applied primarily to prevent severe damage to crops by nematodes during the early stages of growth when they are most vulnerable. Since the nematodes are not killed by non-fumigant nematicides, the extent by which such compounds can reduce damage to the crop will depend on how long the nematodes are inhibited by the nematicides.

A review of factors which influence the control of nematodes with nematicides was given by Whitehead (1978). The review highlights the major factors which can influence the efficacy of non-fumigant nematicides, including soil temperature, organic matter content of the soil, the method of pesticide application (which influences the distribution of the nematicide in the soil) and soil moisture.

Miller and Rich (1974) found that oxamyl was more effective in reducing the population of *Pratylenchus penetrans* in the soil at high temperature whereas carbofuran was more effective at the lower temperature. Conversely, Bunt (1975) found that oxamyl was more effective against *Pratylenchus penetrans* on *Ligustrum ovalifolium* at lower temperatures while phenamiphos was better at the higher temperatures.

The importance of the soil composition in the control of nematodes with non-fumigant nematicides is well documented in the literature. For example, thionazin is known to control *Globodera rostochiensis* better in a peaty loam than silt loam soil (Whitehead *et al.*,1973a) while phenamiphos was better in sandy loam than in a peaty loam or in a silt loam (Whitehead *et al.*,1973b). Bunt, (1975) evaluated the effects of organic matter content of the soil on the efficacy of oxamyl and phenamiphos against *D. dipsaci* on tomato. He found that organic matter content of the soil had little effect on oxamyl, but the nematicidal efficacy of phenamiphos was reduced in organic soils.

The method of application is also an important factor which affects the efficacy of nematicides. The application of dazomet in two equal doses was found to be more effective in the control of *G. rostochiensis* on potato in silt loam soil, than a single application of the equivalent combined dose (Whitehead *et al.*,1973c). The same workers also found that in peaty loam soil and sandy loam soil dazomet killed as many nematodes applied as a single dose after ploughing as two equal doses. Miller and Noegel (1970) have compared the effects of different application methods on the control of *M. incognita* on gardenia plants. They found differences between the various methods investigated, namely bare root dip for 30 minutes, soil drenches, granules broadcast and left uncovered, granules broadcast and covered with one inch of soil and granules thoroughly incorporated into the soil. Soil drenches and granules incorporated into the soil gave the best control. Reddy (1975) reported that the control of *M. incognita* and yields of tobacco from plots treated with fensulphothion, aldicarb and carbofuran at 1kg ai/ha, by spot application were as high as those from plots treated with 4kg ai/ha by row treatment.

The above work suggests that generalizations cannot be made concerning the effects of various factors on the efficacy of non-fumigant nematicides and that factors affecting individual nematicides should be ascertained.

The studies reported in this section set out to evaluate the influence of soil temperature, organic matter content of the soil, and different methods of application on the efficacy of carbofuran against *M. incognita*.

7.1 INFLUENCE OF SOIL TEMPERATURE ON THE EFFICACY OF CARBOFURAN AGAINST *M. INCOGNITA*.

The following experiment was designed to evaluate the effects of soil temperature on the persistence of nematicidal activity of carbofuran against *M. incognita* on eggplant.

MATERIALS AND METHODS

Twenty eight plastic pots, 22.5cm in diameter, were each filled with 5kg of heat-sterilized soil. Carbofuran was added to half of these pots at the rate of 8kg ai/ha and NPK fertilizer at 9.14g/pot. A three week old seedling of eggplant was transplanted into each pot and 2,000 second stage juveniles were added. Fourteen pots (seven treated with carbofuran and seven untreated) were arranged in a fully randomized design on a heated greenhouse bench. This experiment was carried out between the months of October - November when the soil temperature fluctuated between 18^{o} - $20^{o}C$. The remaining fourteen pots were arranged in a fully randomized design in a constant temperature room (soil temperature range - 27^{o} - $30^{o}C$). The pots were watered whenever necessary.

Two weeks after inoculation, three plants from both treated and untreated soil were harvested from pots at each temperature. The root systems of these plants were weighed, the roots cut into small pieces, stained in 0.1% (w/v) cotton blue in lactophenol, macerated in a blender and the number of nematodes per root system estimated. The remaining plants were left until six weeks after inoculation when the experiment was terminated. Fresh weight of shoots and roots were taken and the number of galls per root system counted.

Table 7.2.1 Influence of carbofuran on invasion of eggplant roots by *M. incognita* at two different temperature regimes. (Data taken 2 weeks after inoculation of nematodes)

Treatment	Temperature regime	Fresh weight of roots	No. of nematodes/root system	% reduction in invasion
Soil treated with carbofuran	$27^{o} - 30^{o}C$	5.40[1]	180.7	54.2
Untreated soil	$27^{o} - 30^{o}C$	5.76	395.3	-
Soil treated with carbofuran	$18^{o} - 20^{o}C$	4.60	67.3	82.0
Untreated soil	$18^{o} - 20^{o}C$	4.90	374.0	-
L.S.D. at 5%		N.S.	24.4	
L.S.D. at 1%		N.S.	35.5	

1) Means of 3 replicates

Table 7.2.2 Influence of carbofuran on the growth and root galling of eggplant infected with *M. incognita* at two different temperature regimes (6 weeks after inoculation of nematodes)

Treatment	Temperature regime	Fresh weight of tops	% Increase over untreated	Fresh wt of roots	No. of galls/ root system	% reduction in galling
Soil treated with carbofuran	27° - 30°C	56.20 [1)]	34.12	12.05	502.0	50.88
Untreated soil	27° - 30°C	41.90	-	15.20	1022.0	-
Soil treated with carbofuran	18° - 20°C	52.15	79.20	10.60	235.0	70.70
Untreated soil	18° - 20°C	29.10	-	13.18	804.0	-
L.S.D. at 5%		8.20		2.24	116.5	
L.S.D. at 1%		11.20		3.14	163.4	

1) Means of 4 replicates

RESULTS

At two weeks after inoculation, the application of carbofuran significantly ($p < 0.01$) reduced root invasion by 82% at the lower temperature regime but by 54.2% at the higher temperature regime. (Table 7.2.1). The interaction, nematicide x temperature was significant ($p < 0.05$) for nematode invasion.

At the termination of the experiment six weeks after inoculation, the application of carbofuran significantly ($p < 0.01$) increased top weight of plants by 79.2% at the lower temperature but by 34.1% at the higher temperature (Table 7.2.2). The number of galls per root system was also significantly ($p < 0.01$) reduced by 70.7% and 50.9% at the lower and high temperature regimes, respectively. (Table 7.2.2).

7.2 INFLUENCE OF ORGANIC MATTER CONTENT ON THE EFFICACY OF CARBOFURAN AGAINST *M. INCOGNITA*

The application of organic manures is recommended for the production of vegetables in the tropics (Purseglove, 1968), to supply additional nutrients to the plants and improve soil structure. The increase in the organic matter content of the soil could influence the efficacy of applied non-fumigant nematicides. There is complete paucity of information on the influence of organic matter content of the soil on the efficacy of carbofuran in the control of *M. incognita*.

The following experiment sets out to evaluate the influence of organic matter content of the soil on the efficacy of carbofuran against *M. incognita* attacking eggplant.

MATERIALS AND METHODS

Plastic pots (22.5cm in diameter) were each filled with 5kg of heat-sterilized sand or sand mixed with peat to give 25% and 50% (w/w) organic matter. Fourteen pots were used for each soil type. Half of the pots (7) for each soil type was treated with carbofuran at 8kg ai/ha while the remaining seven pots were untreated. NPK fertilizer (9.14g per pot) was added to each pot before three-week old seedlings of eggplant were transplanted singly into each pot. Soil of each plant was inoculated with 2,000 second stage juveniles. Pots were arranged in the greenhouse in a fully randomized design, and watered when necessary (temperature range 15 - 38^{o}C).

Two weeks after inoculation, three plants were taken from both carbofuran treated and untreated soils at the different levels of organic matter. The roots were washed thoroughly, weighed and the number of nematodes per root system estimated as described previously.

Table 7.3.1 Influence of carbofuran on invasion of eggplant roots by *M. incognita* in soils with different organic matter content. (Data taken 2 weeks after inoculation).

Treatment	No. of nematodes/root system[1)]	% reduction in invasion
Sand		
Treated with carbofuran	119.7	72.07
Untreated	428.7	-
25% organic matter		
Treated with carbofuran	128.7	69.08
Untreated	416.3	-
50% organic matter		
Treated with carbofuran	268.7	13.24
Untreated	309.7	-
L.S.D. at 5%	31.87	
L.S.D. at 1%	44.69	

1) Means of 3 replicates

Table 7.3.2 Influence of carbofuran on the growth and root galling of eggplant infected with *M. incognita* in soils with different organic matter content.

Treatments	Fresh weight of tops 1)	% increase over untreated	Fresh weight of roots	No. of galls/root system	% reduction in galling
Sand:					
Treated with carbofuran	63.90	53.60	10.30	346.0	69.15
Untreated	41.60	-	13.30	1121.5	-
25% organic matter					
Treated with carbofuran	60.80	30.33	11.20	425.0	56.89
Untreated	46.65	-	14.20	986.0	-
50% organic matter					
Treated with carbofuran	56.80	14.52	12.90	724.0	8.68
Untreated	49.60	-	15.10	792.8	-
L.S.D. at 5%	8.24		1.7	116.8	
L.S.D. at 1%	11.20		2.6	159.3	

1) Means of 4 replicates

The remaining plants were allowed to grow until six weeks after inoculation. The fresh weight of tops and roots was then taken and the number of galls per root system estimated.

RESULTS

At two weeks, the application of carbofuran gave a 72.1% reduction in sand, 69.1% in soil with 25% (w/w) organic matter but only 13.2% in soil with 50% (w/w) organic matter (Table 7.3.1).

The interaction, nematicide x soil type was significant for nematode invasion. ($p<0.01$).

At six weeks after inoculation, the application of carbofuran significantly ($p < 0.01$) increased the fresh top weight by 53.6% and 30.3% in sand and soil containing 25% (w/w) organic matter respectively, but in soil containing 50% (w/w) organic there was only an increase of 14.5% and this was not significant ($p > 0.05$) (Table 7.3.2).

The number of galls per root system was significantly ($p < 0.01$) reduced by 69.1% and 56.9% in sand and soil containing 25% (w/w) organic matter respectively, following the application of carbofuran but in soil containing 50% (w/w) organic matter a reduction of 8.7% was found and this was not significant ($p > 0.05$).

7.3 EFFECTS OF SINGLE AND MULTIPLE APPLICATIONS OF CARBOFURAN ON CONTROL OF *M. INCOGNITA*

Various methods of applying nematicides have been compared to determine the most effective method for particular pest problems (Hague *et al.*, 1964, Whitehead *et al.*, 1975a; Whitehead *et al.*, 1975b; Whitehead *et al.*, 1979). The longer the persistence of activity of the nematicide, the greater the reduction in damage caused by the nematodes to the plant.

One of the ways of achieving this is to apply the nematicide in two equal doses (Whitehead *et al.*, 1973c) or by other multiple applications.

The time the second application should be made is crucial because the problem of phytotoxicity could offset the beneficial effects resulting from better nematode control.

The following experiment was designed to evaluate various methods of application on the efficacy of carbofuran on *M. incognita* on eggplant.

MATERIALS AND METHODS

Soil heavily infested with *M. incognita* was mixed with appropriate amounts of heat-sterilized soil to give a population density of 150 second stage juveniles of *M. incognita* per 200g of soil (3,750 second stage juveniles per pot). Plastic pots (22.5cm in diameter) were each filled with 5kg of infested soil and carbofuran granules were added to the pots at the rate of 8kg a.i./ha (200mg of Furadan 10G per pot) as a single dose at transplanting and in two equal doses (4kg a.i./ha) (100mg at transplanting and 100mg two weeks later). In two other treatments additional carbofuran was added at 4kg a.i./ha to soil in

Plate 4 Phytotoxicity symptoms on leaves of eggplant given a second treatment of carbofuran 2 weeks after transplanting

The second treatment of carbofuran given 3 weeks after transplanting

TABLE 10 EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF CARROT INFECTED WITH *M. INCOGNITA*

FINAL SOIL POPULATION OF L_2 JUVENILES/200g OF SOIL

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	185	225	140	115	166.3
2. Carbofuran + Nematodes	70	55	75	55	71.3
3. Fertilizer + Carbofuran + Nematodes	95	105	85	155	85.0
4. None + Nematodes	85	100	70	135	95.5

APPENDIX C

TABLE 11 EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF CARROT INFECTED WITH *M. INCOGNITA*

SCORES FOR ROOT DEFORMATION

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	3.0	3.0	3.0	3.0	3.0
2. Carbofuran + Nematodes	2.0	2.0	2.0	2.0	2.0
3. Fertilizer + Carbofuran + Nematodes	2.0	2.0	2.0	2.0	2.0
4. None + Nematodes	3.0	3.0	3.0	3.0	3.0

APPENDIX D

TABLE 1 MECHANISM OF ACTION OF FERTILIZERS ON *M. INCOGNITA*

EFFECTS OF FERTILIZERS ON ROOT INVASION

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
No Fertilizer applied	200	226	210	182	204.5
Normal rate (18.29g/pot)	196	176	210	190	193
1.5 x normal rate	183	190	200	173	186.5
2 x normal rate	156	163	186	170	168.8

APPENDIX D

TABLE 2 MECHANISM OF ACTION OF FERTILIZERS ON M.INCOGNITA

INCREASE IN FRESH SHOOT WEIGHT WITH TIME

TREATMENTS	REPLICATES	3 Weeks	6 Weeks
Fertilizer + Nematodes	1	25.6	58.2
	2	31.2	64.0
	3	20.1	53.2
	4	28.7	63.4
	Mean	26.4	59.7
Fertilizer - Nematodes	1	26.9	78.9
	2	30.4	70.7
	3	35.0	76.1
	4	28.9	73.7
	Mean	30.3	74.8
None + Nematodes	1	11.9	18.5
	2	15.2	16.9
	3	17.8	23.2
	4	10.7	14.2
	Mean	13.9	18.2
None - Nematodes	1	20.8	46.4
	2	14.9	51.3
	3	17.5	42.9
	4	23.2	56.2
	Mean	19.1	49.2

APPENDIX D

TABLE 3 MECHANISM OF ACTION OF FERTILIZERS ON M.INCOGNITA

INCREASE IN DRY SHOOT WEIGHT WITH TIME

TREATMENTS	REPLICATES	3 Weeks	6 Weeks
Fertilizer + Nematodes	1	4.31	9.49
	2	5.13	10.87
	3	3.44	8.91
	4	4.69	10.61
	Mean	4.39	9.97
Fertilizer - Nematodes	1	4.50	13.26
	2	5.03	11.83
	3	5.78	12.71
	4	4.39	12.33
	Mean	4.92	12.65
None + Nematodes	1	2.01	3.10
	2	2.61	2.91
	3	3.11	3.76
	4	1.72	2.39
	Mean	2.36	3.02
None - Nematodes	1	3.39	7.81
	2	2.59	8.49
	3	2.84	7.21
	4	3.69	9.42
	Mean	3.12	8.23

APPENDIX D

TABLE 4 MECHANISM OF ACTION OF FERTILIZERS ON *M. INCOGNITA*

INCREASE IN FRESH ROOT WEIGHT WITH TIME

TREATMENTS	REPLICATES	3 Weeks	6 Weeks	9 Weeks
Fertilizer + Nematodes	1	11.4	26.9	48.4
	2	13.8	21.5	39.9
	3	10.8	30.2	36.8
	4	15.2	28.2	47.1
	Mean	12.8	26.7	43.0
Fertilizer - Nematodes	1	9.8	18.6	29.6
	2	11.2	20.8	36.9
	3	10.4	18.5	30.8
	4	9.0	24.2	32.2
	Mean	10.1	20.5	32.4
None + Nematodes	1	7.4	15.2	14.9
	2	5.9	10.7	22.9
	3	6.6	12.9	20.6
	4	5.9	14.0	17.6
	Mean	6.4	13.2	18.9
None - Nematodes	1	6.2	11.6	16.2
	2	5.3	10.9	17.3
	3	4.9	13.8	18.2
	4	6.4	10.1	13.9
	Mean	5.7	11.6	16.4

APPENDIX D

TABLE 5 MECHANISM OF ACTION OF FERTILIZERS ON M.INCOGNITA

INCREASE IN ROOT LENGTH WITH TIME

TREATMENTS	REPLICATES	3 Weeks	6 Weeks	9 Weeks
Fertilizer + Nematodes	1	18.9	24.9	30.8
	2	20.7	29.8	38.2
	3	18.6	26.8	31.2
	4	23.4	30.1	32.6
	Mean	20.4	27.9	33.2
Fertilizer - Nematodes	1	22.8	36.9	36.2
	2	20.9	27.6	40.5
	3	24.9	33.2	42.8
	4	22.6	31.9	38.9
	Mean	22.8	32.4	39.6
None + Nematodes	1	15.5	10.6	15.9
	2	9.9	18.0	12.2
	3	13.2	12.1	18.8
	4	12.6	14.9	10.7
	Mean	12.8	13.9	14.4
None - Nematodes	1	18.9	24.8	26.9
	2	14.2	16.9	32.9
	3	21.8	28.2	23.7
	4	12.7	19.7	34.9
	Mean	16.9	22.4	29.6

APPENDIX D

TABLE 6 MECHANISM OF ACTION OF FERTILIZERS ON *M. INCOGNITA*

INCREASE IN NUMBER OF GALLS PER ROOT SYSTEM WITH TIME

TREATMENTS	REPLICATES	3 Weeks	6 Weeks
Fertilizer + Nematodes	1	592	2313
	2	621	1892
	3	604	2083
	4	653	2088
	Mean	617	2086
None + Nematodes	1	584	1763
	2	619	1498
	3	567	1625
	4	554	1652
	Mean	581	1634

INCREASE IN NUMBER OF GALLS PER GRAM OF ROOT WITH TIME

TREATMENTS	REPLICATES	3 Weeks	6 Weeks
Fertilizer + Nematodes	1	52	86
	2	45	88
	3	56	69
	4	43	73
	Mean	49	79
None + Nematodes	1	79	116
	2	105	140
	3	86	126
	4	94	118
	Mean	91	125

APPENDIX D

TABLE 7 MECHANISM OF ACTION OF FERTILIZERS ON M.INCOGNITA

INCREASE IN PROPORTION OF ROOTS GALLED WITH TIME (SCORES)

TREATMENTS	REPLICATES	3 Weeks	6 Weeks	9 Weeks
Fertilizer + Nematodes	1	1.0	2.0	4.0
	2	1.0	3.0	4.0
	3	1.0	2.0	4.0
	4	1.0	3.0	4.0
	Mean	1.0	2.5	4.0
None + Nematodes	1	2.0	4.0	5.0
	2	2.0	4.0	5.0
	3	2.0	4.0	5.0
	4	1.0	3.0	5.0
	Mean	1.75	3.75	5.0

APPENDIX D

TABLE 8 MECHANISM OF ACTION OF FERTILIZERS ON *M. INCOGNITA*

IN VITRO EFFECTS OF FERTILIZERS ON *M. INCOGNITA* JUVENILES - NO. OF ACTIVE NEMATODES

		CONCENTRATIONS OF FERTILIZER				
TIME	REPLICATES	0	10 ppm	100 ppm	1000 ppm	10,000 ppm
6	1	10	10	10	10	10
	2	10	10	10	10	10
	3	10	10	10	10	10
24	1	10	10	10	6	3
	2	10	10	10	4	5
	3	10	10	10	5	3
48	1	10	10	10	5	2
	2	10	10	10	4	3
	3	10	10	10	3	2
72	1	10	10	8	3	2
	2	10	10	7	3	1
	3	10	10	8	3	4
96	1	10	10	7	2	0
	2	10	10	7	3	0
	3	10	10	8	2	0
7 day	1	7	9	7	2	0
	2	8	8	6	1	0
	3	8	7	6	2	0

APPENDIX E

TABLE 1 MECHANISM OF ACTION OF CARBOFURAN

EFFECT OF CARBOFURAN AND 3 HYDROXYCARBOFURAN ON ROOT INVASION BY M.INCOGNITA

	TREATMENTS	REPLICATES				MEAN
		1	2	3	4	
CARBOFURAN	Control I 1% Phostrogen	65	60	55	79	64.7
	Control II 1% Acetone	56	63	68	58	61.2
	2ug/g of sand	15	19	22	20	19.0
	4ug/g of sand	11	14	9	11	11.2
	8ug/g of sand	3	4	6	7	5.0
3 HYDROXY-CARBOFURAN	2ug/g of sand	53	69	55	68	61.2
	4ug/g of sand	62	61	55	68	61.5
	8ug/g of sand	55	67	59	47	57.0

APPENDIX E

TABLE 2 MECHANISM OF ACTION OF CARBOFURAN

INFLUENCE OF CONCENTRATION AND EXPOSURE TIME ON INVASION

1 DAY EXPOSURE TIME

CONCENTRATION OF CARBOFURAN	REPLICATES			MEAN
	1	2	3	
Control	67	56	62	61.6
2 ppm	76	68	57	67.0
4 ppm	57	68	53	59.3
8 ppm	59	50	62	57.0

7 DAY EXPOSURE TIME

CONCENTRATION OF CARBOFURAN	REPLICATES			MEAN
	1	2	3	
0	59	48	53	53.3
2 ppm	37	41	48	42.0
4 ppm	38	48	50	45.3
8 ppm	46	39	39	41.3

APPENDIX E

TABLE 3 MECHANISM OF ACTION OF CARBOFURAN

BARE ROOT DIPPING TEST

2 DAYS AFTER REPLANTING

TREATMENTS	REPLICATES			MEAN
	1	2	3	
Control	375	321	404	366.6
2 ppm	330	420	326	358.6
4 ppm	350	236	227	271.0
8 ppm	250	334	329	304.3

9 DAYS AFTER REPLANTING

TREATMENTS	REPLICATES			MEAN
	1	2	3	
Control	294	382	297	324.3
2 ppm	328	385	306	339.6
4 ppm	254	297	332	294.3
8 ppm	296	218	329	281.0

APPENDIX E

TABLE 4 MECHANISM OF ACTION OF CARBOFURAN

MIGRATION OF SECOND STAGE JUVENILES THROUGH A SAND COLUMN

TREATMENTS	REPLICATES			MEAN
	1	2	3	
0 ppm	37	34	32	34.3
1 ppm	35	35	38	36.0
2 ppm	29	34	32	31.6
4 ppm	24	19	20	21.0
8 ppm	14	16	10	13.3

APPENDIX E

TABLE 5 EFFECT OF CARBOFURAN ON HATCHING

(5a) IN PESTICIDE - NO. OF JUVENILES

TREATMENTS	REPLICATES			MEAN
	1	2	3	
0 (Control)	586	499	663	582.6
Carbofuran at 1 ppm	624	478	605	569.0
" " 2 ppm	484	229	342	375.0
" " 4 ppm	338	265	340	314.3
" " 8 ppm	166	126	202	164.6

(5b) IN WATER - NO. OF JUVENILES

TREATMENTS	REPLICATES			MEAN
	1	2	3	
0 (Control)	345	524	409	396.6
Carbofuran at 1 ppm	489	350	604	481.0
" " 2 ppm	535	703	596	611.3
" " 4 ppm	772	845	696	771.0
" " 8 ppm	882	652	952	828.6

(5c) TOTAL HATCH - NO. OF JUVENILES

TREATMENTS	REPLICATES		
	1	2	3
0 (Control)	582.6	392.6	975.2
Carbofuran at 1 ppm	569.0	481.0	1050.0
" " 2 ppm	378.3	611.3	989.6
" " 4 ppm	314.3	771.0	1085.3
" " 8 ppm	164.6	828.6	993.2

APPENDIX E

MECHANISM OF ACTION OF CARBOFURAN

SYSTEMIC EFFECT OF CARBOFURAN ON *M. INCOGNITA*

TABLE 1 THREE DAYS ALLOWED FOR INVASION (NO. OF JUVENILES AND FEMALES PER ROOT SYSTEM)

TREATMENTS	Rate of Carbofuran kg a.i/ha	No. of Females	No. of J_2-J_4 Juveniles
1. Plants from untreated into treated soil	4	49, 48, 41	24, 19, 23
2. "	8	43, 53, 45	27, 23, 34
3. "	16	51, 42, 47	26, 18, 25
4. Plants from untreated into untreated soil	0	43, 53, 39	30, 21, 26

APPENDIX F

TABLE 2 SEVEN DAYS ALLOWED FOR INVASION (NO. OF JUVENILES AND FEMALES PER ROOT SYSTEM)

TREATMENTS	Rate of Carbofuran kg a.i/ha^{-1}	No. of Females	No. of J_2-J_4 Juveniles
1. Plants from untreated into treated soil	4	66, 70, 63	26, 23, 26
2. "	8	80, 76, 76	30, 36, 33
3. "	16	86, 83, 90	33, 40, 30
4. Plants from untreated into untreated soil	0	83, 76, 86	20, 26, 33

APPENDIX F

FACTORS INFLUENCING THE EFFECTS OF FERTILIZERS ON *M. INCOGNITA*

TABLE 1 EFFECTS OF FERTILIZER RATES

FRESH WEIGHT OF TOPS

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	97.9	81.6	79.3	94.8
2 Fertilizers - Nematodes	91.8	118.7	102.6	94.1
2 Fertilizers + Nematodes	77.9	86.9	79.6	97.2
2 Fertilizers - Nematodes	96.2	101.6	112.7	91.9
Fertilizer + Nematodes	76.1	61.8	66.8	72.9
Fertilizer - Nematodes	99.6	84.9	102.6	95.3
Fertilizer + Carbofuran + Nematodes	104.2	86.9	97.7	81.6
Fertilizer + Carbofuran - Nematodes	101.5	110.8	103.6	96.9
None + Nematodes	10.9	6.4	8.1	13.0
None - Nematodes	64.8	50.9	63.7	54.2

APPENDIX F

TABLE 2 NO OF DEVELOPING FRUITS

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
2 Fertilizers + Nematodes	3.0	2.0	3.0	3.0	2.8
2 Fertilizers - Nematodes	4.0	5.0	5.0	3.0	4.2
2 Fertilizers + Nematodes	4.0	4.0	3.0	5.0	4.0
2 Fertilizers - Nematodes	5.0	4.0	3.0	4.0	4.0
Fertilizer + Nematodes	2.0	1.0	2.0	1.0	1.5
Fertilizer - Nematodes	3.0	2.0	3.0	4.0	3.0
Fertilizer + Carbofuran + Nematodes	4.0	5.0	4.0	4.0	4.2
Fertilizer + Carbofuran - Nematodes	6.0	4.0	3.0	6.0	4.7
None + Nematodes	0	0	0	0	0
None - Nematodes	0	0	1	1	0.5

TABLE 3 FRESH ROOT WEIGHTS

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	53.2	46.8	45.9	56.5
2 Fertilizers - Nematodes	39.7	43.1	38.9	39.1
2 Fertilizers + Nematodes	59.6	66.9	62.7	57.2
2 Fertilizers - Nematodes	56.2	46.2	49.9	48.1
Fertilizer + Nematodes	46.2	38.2	40.7	45.2
Fertilizer - Nematodes	30.6	39.8	30.7	31.9
Fertilizer + Carbofuran - Nematodes	41.1	39.6	32.8	31.9
Fertilizer + Carbofuran - Nematodes	38.2	30.6	28.6	36.2
None + Nematodes	8.4	7.8	6.8	9.2
None - Nematodes	13.8	11.9	14.1	15.8

APPENDIX F

TABLE 4 RATINGS FOR ROOT GALLING AND % OF ROOTS GALLED

TREATMENTS	ROOT GALLING REPLICATES				% OF ROOTS GALLED REPLICATES			
	1	2	3	4	1	2	3	4
2 Fertilizers + Nematodes	5.0	5.0	5.0	5.0	3.0	3.0	3.0	3.0
2 Fertilizers + Nematodes	5.0	5.0	5.0	5.0	3.0	3.0	3.0	3.0
Fertilizer + Nematodes	5.0	5.0	5.0	5.0	4.0	4.0	4.0	4.0
Fertilizer + Carbofuran + Nematodes	4.0	4.0	4.0	4.0	3.0	3.0	3.0	3.0
None + Nematodes	6.0	6.0	6.0	6.0	5.0	5.0	5.0	5.0

APPENDIX F

TABLE 5 FINAL SOIL POPULATIONS OF JUVENILES PER 200g OF SOIL

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	2650	1500	2455	1595
2 Fertilizers + Nematodes	3100	2050	2255	2195
Fertilizer + Nematodes	1895	1150	1925	1630
Fertilizer + Carbofuran + Nematodes	1855	1600	2150	1645
None + Nematodes	595	390	725	450

APPENDIX G1

HIGH SOIL POPULATION DENSITY

TABLE 1 FRESH WEIGHT OF TOPS

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	71.3	62.9	58.6	68.4
2 Fertilizers - Nematodes	115.7	92.7	102.4	119.6
Fertilizer + Nematodes	52.7	41.8	51.2	48.6
Fertilizer - Nematodes	93.4	84.2	99.2	89.6
None + Nematodes	0.0	0.0	0.0	0.0
None - Nematodes	49.3	58.6	43.1	50.6

TABLE 2 RATINGS FOR ROOT GALLING AND % OF ROOTS GALLED

TREATMENTS	ROOT GALLING REPLICATES				% OF ROOTS GALLED REPLICATES			
	1	2	3	4	1	2	3	4
2 Fertilizers + Nematodes	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Fertilizer + Nematodes	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
None + Nematodes	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

APPENDIX G_1

TABLE 3 FINAL SOIL POPULATIONS OF L_2 JUVENILES/200g OF SOIL

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	3270	2460	4320	3950
Fertilizer + Nematodes	2695	2955	3120	3620
None + Nematodes	0	0	0	0

APPENDIX G_1

TABLE 4 FRESH ROOT WEIGHTS

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	43.7	49.9	46.1	45.9
2 Fertilizers - Nematodes	40.6	43.1	32.9	37.0
Fertilizer + Nematodes	39.6	29.8	36.2	28.8
Fertilizer - Nematodes	33.6	30.4	27.8	33.0
None + Nematodes	0	0	0	0
None - Nematodes	16.0	11.9	14.3	16.2

TABLE 5 NO. OF DEVELOPING FRUITS

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	0	0	0	0
2 Fertilizers - Nematodes	4	4	5	3
Fertilizer + Nematodes	0	0	0	0
Fertilizer - Nematodes	2	3	2	4
None + Nematodes	0	0	0	0
None + Nematodes	0	0	0	0

APPENDIX G_2

INFLUENCE OF PLANT AGE

TABLE 1 SHOOT WEIGHT (ONE WEEK OLD SEEDLINGS)

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Fertilizer + Nematodes	24.2	19.3	26.4	16.9	21.70
Fertilizer - Nematodes	39.2	43.7	29.6	38.7	37.80
None + Nematodes	9.5	5.7	7.1	8.9	7.80
None - Nematodes	29.2	21.6	28.9	23.7	25.85

APPENDIX G_2

TABLE 2 FRESH ROOT WEIGHT (ONE WEEK OLD SEEDLINGS)

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Fertilizer + Nematodes	12.6	8.4	10.3	9.1	10.10
Fertilizer - Nematodes	10.0	8.6	8.4	10.4	9.35
None + Nematodes	3.3	4.2	3.4	3.9	3.70
None - Nematodes	4.6	6.6	4.9	5.1	5.20

APPENDIX G_2

TABLE 3 FRESH WEIGHT OF TOPS (FOUR WEEK OLD SEEDLINGS)

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Fertilizer + Nematodes	89.6	96.2	81.9	90.7	89.60
Fertilizer - Nematodes	75.6	98.6	79.6	84.9	84.67
None + Nematodes	31.8	24.8	39.6	34.2	32.60
None - Nematodes	49.8	54.2	43.2	59.6	51.70

APPENDIX G_2

TABLE 4 FRESH WEIGHT OF ROOTS (FOUR WEEK OLD SEEDLINGS)

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Fertilizer + Nematodes	48.9	40.2	46.4	38.9	43.60
Fertilizer - Nematodes	33.3	38.4	30.6	34.9	34.30
None + Nematodes	14.9	13.2	16.7	16.7	15.38
None - Nematodes	16.9	11.9	14.2	12.6	13.90

EFFECT OF VARIOUS NPK COMBINATIONS

APPENDIX G_3

TABLE 1 FRESH TOP WEIGHTS

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
$N_- P_1 K_1$ + Nematodes	12.7	9.8	15.4	10.9	12.20
$N_- P_1 K_1$ - Nematodes	33.4	39.3	31.9	28.2	33.20
$N_1 P_- K_1$ + Nematodes	18.4	20.6	17.9	13.9	17.70
$N_1 P_- K_1$ - Nematodes	37.8	34.7	39.3	32.6	36.10
$N_1 P_1 K_-$ + Nematodes	31.9	21.9	34.9	24.5	28.30
$N_1 P_1 K_-$ - Nematodes	39.2	47.8	40.2	49.6	44.20
$N_1 P_1 K_1$ + Nematodes	38.4	48.9	44.8	33.3	40.10
$N_1 P_1 K_1$ - Nematodes	47.9	60.5	58.3	43.7	52.60
$N_2 P_1 K_1$ + Nematodes	44.5	39.6	36.1	47.2	41.85
$N_2 P_1 K_1$ - Nematodes	53.0	49.2	46.1	58.9	51.80
$N_1 P_2 K_1$ + Nematodes	37.2	45.3	32.9	42.6	39.50
$N_1 P_2 K_1$ - Nematodes	55.9	46.2	60.3	51.2	53.40
$N_1 P_1 K_2$ + Nematodes	36.9	43.7	39.7	38.9	39.80
$N_1 P_1 K_2$ - Nematodes	48.7	46.2	57.3	48.6	50.20

TABLE 2 FRESH ROOT WEIGHTS

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
$N- P_1 K_1$ + Nematodes	4.8	6.9	5.7	4.2	6.20
$N- P_1 K_1$ - Nematodes	7.2	5.1	5.5	7.0	5.40
$N_1 P- K_1$ + Nematodes	6.4	6.8	8.2	7.2	8.90
$N_1 P- K_1$ - Nematodes	9.8	8.4	7.3	10.1	7.15
$N_1 P_1 K-$ + Nematodes	9.1	7.7	8.8	8.0	8.40
$N_1 P_1 K-$ - Nematodes	7.9	6.9	7.5	6.5	7.20
$N_1 P_1 K_1$ + Nematodes	12.5	13.6	10.2	12.9	12.30
$N_1 P_1 K_1$ - Nematodes	8.9	12.6	11.2	8.1	10.20
$N_2 P_1 K_1$ + Nematodes	11.6	9.3	11.0	8.9	11.85
$N_2 P_1 K_1$ - Nematodes	8.9	10.8	10.4	8.3	10.20
$N_1 P_2 K_1$ + Nematodes	14.2	11.6	16.5	10.9	13.30
$N_1 P_2 K_1$ + Nematodes	10.6	11.9	13.2	9.1	11.20
$N_1 P_1 K_2$ + Nematodes	11.2	15.9	13.6	10.9	12.90
$N_1 P_1 K_2$ - Nematodes	13.4	9.7	12.1	8.4	10.90

APPENDIX G_3

TABLE 3 RATINGS FOR ROOT GALLING AND PERCENTAGE OF ROOTS GALLED

TREATMENTS	RATINGS FOR ROOT GALLING REPLICATES				% OF ROOTS GALLED REPLICATES			
	1	2	3	4	1	2	3	4
$N -P_1 K_1$ + Nematodes	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
$N_1 P -K_1$ + Nematodes	5.0	5.0	5.0	5.0	4.0	5.0	5.0	4.0
$N_1 P_1 K-$ + Nematodes	5.0	5.0	5.0	5.0	4.0	5.0	5.0	5.0
$N_1 P_1 K_1$ + Nematodes	5.0	5.0	5.0	5.0	3.0	3.0	3.0	3.0
$N_2 P_1 K_1$ + Nematodes	5.0	5.0	5.0	5.0	3.0	3.0	3.0	3.0
$N_1 P_2 K_1$ + Nematodes	5.0	5.0	5.0	5.0	3.0	3.0	3.0	3.0
$N_1 P_1 K_2$ + Nematodes	5.0	5.0	5.0	5.0	3.0	3.0	3.0	3.0

pots previously treated with 8kg a.i./ha carbofuran at two and three weeks after transplanting. In each treatment, four-week old seedlings of eggplant were transplanted singly into each pot and NPK fertilizer added at the rate of 9.14g per pot. Pots given fertilizers alone served as control. Each treatment was replicated four times. Details of the various treatments are given in Table 7.4.1. Pots were arranged in a fully randomized design in the greenhouse (temperature range 14-38°C). Eight weeks after transplanting plants were harvested, the fresh weight of tops and roots taken and the number of galls per root system counted.

RESULTS

At the termination of the experiment, the number of galls per root system was significantly ($p < 0.01$) reduced by single and multiple application of carbofuran when compared with control (Table 7.4.1). The greatest reduction in number of galls per root system was obtained when carbofuran was applied in multiple application. A reduction of 86.1% and 82.5% were obtained when the second application of carbofuran was done 2 and 3 weeks after transplanting, respectively. But root galling was reduced by 68.2% when applied as a single dose and 54.9% when applied in split doses.

Fresh top weights were all increased by the application of carbofuran when applied as single and as multiple application ($p < 0.05$). There was no significant ($p > 0.05$) difference between the fresh top weights obtained for plants in carbofuran treated soil, irrespective of application method. (Table 7.4.1). Phytotoxicity symptoms were observed on the leaves of plants in soils given a second application of carbofuran. The symptoms were seen as white necrotic spots on the upper leaf surface and browing of leaf edges. The symptoms were more pronounced when carbofuran

Table 7.4.1 Effect of single and multiple applications of carbofuran on root galling and growth of eggplant infected with *M. incognita*

Treatments	Dosage kg ai/ha	No. of galls per root system	% reduction in root galling	Fresh weight of tops	Fresh weight of roots
Carbofuran applied as a single dose at transplanting	8	962	68.22	82.60	28.38
Carbofuran applied in split doses	4 at transplanting + 4 two weeks later	1035	54.98	77.20	26.93
Extra dose of carbofuran applied two weeks after transplanting	8 at transplanting 4 two weeks later	416	86.26	75.15	20.60
Extra dose of carbofuran applied three weeks after transplanting	8 at transplanting 4 three weeks later	529	82.52	83.70	25.20
Untreated with nematicide (Control)	-	3028	-	61.95	42.65
L.S.D. at 5%		206.4		13.0	4.97
L.S.D. at 1%		285.5		17.9	6.88

was applied again at two weeks after transplanting (see Plate 4).

DISCUSSION

The results have shown that soil temperature is an important factor influencing the efficacy of carbofuran against *M. incognita*. Carbofuran was more effective in preventing the invasion of the roots of eggplant by *M. incognita* at the lower temperature (Table 7.2.1).

The results however, show too that carbofuran can still be used quite effectively at higher temperatures to reduce root-knot damage and increase crop yields. The results obtained here are similar to those of Miller and Rich (1974) who found that carbofuran was more effective against *Pratylenchus penetrans* at the lower temperature. The reduced effectiveness at the higher temperature may be related to faster degradation of carbofuran to non toxic metabolites at this temperature. Caro *et al.* (1973) found that carbofuran persistence in soil increased at low soil temperatures. Talekar *et al.* (1977) also found that the breakdown of carbofuran was accelerated during the hot season.

The results showed clearly that the effect of carbofuran on the nematodes was markedly reduced in soil with 50% w/w organic matter. The poorer control in soil with high organic matter content indicates that carbofuran has a high partition coefficient which according to Bromilow (1973) influences the effectiveness of pesticides in the soil with different organic matter

$$\text{Partition Coefficient } Q = \frac{\text{chemical concentration in soil organic matter}}{\text{chemical concentration in soil water}}$$

The greater the Q the less the proportion of the chemical in soil water and hence the greater the amount absorbed onto the organic matter fraction of the soil. Although the Q value of carbofuran has not been measured, Bromilow (1980), however, gave the predicted Q value to be 31 which is quite high when compared with those of aldicarb (15) and oxamyl (2.4).

The results obtained on the influence of single and multiple application has shown that the efficacy of carbofuran was increased when an extra treatment was given. Although phytotoxicity symptoms were observed growth was not adversely effected. This is of practical significance as it suggests that carbofuran can be effectively used in situations where an extra treatment is needed even after a very short interval. However, differences may exist between crops in relation to how they respond to high concentrations of chemicals. Soil conditions might also be important as Lamberti (1971a) has reported that carbofuran is toxic to tomato in alkaline soils. Tolerance to phytotoxic effect as found might also be related to the conditions under which this experiment was carried out as watering was done daily. In the field where rainfall is erratic, caution must be taken.

GENERAL DISCUSSION AND CONCLUSIONS

This project was initiated to study the effects of fertilizers and the use of chemical control agents on the pathogenicity of *M. incognita* in relation to vegetable production, particularly as it applies to conditions in the tropics. In particular, it was hoped that from the results it would be possible to determine whether fertilizers could be included in a management programme designed to minimize crop losses due to root-knot nematodes in my country, Nigeria.

It was considered necessary to evaluate the application of fertilizers alone as a means of alleviating the damage caused by root-knot nematodes to vegetables as other cultural practices such as crop rotation, fallowing and flooding are not always economically practicable in Nigeria. Because of the polyphagous nature of root-knot nematodes, suitable high-value cash crops which can be grown in place of the highly profitable vegetable crops have yet to be identified. Fallowing under irrigation is impracticable as there are limitations in availability of irrigated areas while in upland areas the presence of weed hosts helps to maintain populations of the nematodes under fallow conditions. Flooding is undesirable as it has adverse effects on soil structure and the costs of maintaining flooded fields and drainage would be too high.

Because of the high costs of nematicides in Nigeria, carbofuran, a broad-spectrum organocarbamate pesticide with insecticidal as well as nematicidal properties was selected for study as its use on vegetables would reduce production costs by eliminating or minimizing further application of insecticides. Carbofuran was chosen in preference to other non-fumigant nematicides with similar properties such as aldicarb (Temik), oxamyl (Vydate) and phenamiphos (Nemacur) because of its low dermal toxicity which reduces the risk of poisoning.

Since this work was directed at the control of *M. incognita* on vegetables in Nigeria, attempts were made to simulate the conditions found there during the growing season. All experiments were done in the summer in heated glass houses when the soil temperature in pots was in the range found during the growing season in Nigeria. Sandy loam soils with low organic matter content and with pH of between 5 - 7 were used. Seinhorst (1979) considered the varying effects of nematodes on plants grown in pots and under field conditions to be due mainly to differences in the sources of inoculum In the present work second stage juveniles were used as these are the source of infection after the dry season in Nigeria. These nematodes survive the dry season in the egg stage and hatch soon after the first rains prior to planting (Odihirin, 1976). Inoculation was done in such a way as to ensure even distribution of the nematodes around the plants, as this could be another source of variation from field conditions.

It was shown that the applications of NPK fertilizers at the recommended rate markedly lessened the damaging effects of *M. incognita* on egg plant. This agrees in principle with the results of Oteifa (1952); Vazquez (1967); Ross (1969); Smolik and Malek (1973), who have also found that the application of certain fertilizers can be used to lessen the detrimental effects of plant parasitic nematodes to plants. The growth of lettuce and carrot was enhanced by the application of fertilizers but the amount of crop lost due to the nematodes was unaltered.

The ineffectiveness of fertilizer application in reducing the damage caused by *M. incognita* to lettuce and carrot can be related to the method by which they are cultivated i.e. by direct seeding. The importance of the method of cultivation as a factor influencing the effects of fertilizers on damage caused by nematodes to plants has not been previously reported.

The level and proportion in which NPK fertilizers were combined, the level of infestation and plant age, were identified as other factors influencing the effects of fertilizers on damage to egg plant. Plants inoculated with 3,000 second stage juveniles and given twice the rate of NPK fertilizers as a split application produced the same number of developing fruits as those found on uninfected plants. However, no fruits were produced on infected plants at high soil infestation level (15,000 second stage juveniles/plant) even when twice the rate of fertilizers was given. The application of fertilizers that have no nematicidal properties is unlikely to be useful in vegetable production in soils where continuous infection of newly formed roots will occur.

Deficiency of N, P and K in sand resulted in greater damage to egg plant when compared to that found when plants were grown in sand given the complete nutrients. The greatest damage to egg plant occurred when nitrogen was deficient which agrees with the findings of Shafiee and Jenkins (1962) in relation to the damage to pepper by *Pratylenchus penetrans*. Deficiency of any of the nutrients but particularly nitrogen, will result in severe crop losses due to *M. incognita* on egg plant. Increasing the levels of nitrogen and potassium resulted in greater tolerance of the plants to the nematodes. Bessey (1911), Oteifa et al (1965) have similarly found that increasing the amounts of potassium further increased the tolerance of plants to nematodes.

The reduction in the damaging effects of *M. incognita* on egg plant after fertilizer application was found to be due to improved tolerance of the plants. This is in agreement with the findings of other workers on the tolerance of plants to nematode damage following the application of fertilizers (Bessey, 1911; Oteifa et al 1965, Vazquez, 1967). It was shown that the improved tolerance of infected plants in soils given fertilizers was related to their enhanced capacity to compensate for damaged roots as was indicated by the greater percentage of roots which were not galled compared to those of plants in unfertilized soils when assessments were made at three, six and nine weeks after inoculation. The mechanism of tolerance of plants to damage by root-knot nematodes when given fertilizers has not been previously elucidated. Lateral root proliferation, which occurs when root axis extension is disrupted by nematodes (Price, 1979), probably accounts for most of the roots not being galled although some of the increase in healthy roots might be due to primary roots capable of growing away from the source of infection. Investigations should be done on the effect of fertilizer application on the post invasion development of root systems of plants infected with root-knot nematodes to elucidate this.

It was shown that the number of galls per root system and per gram of root of plants in soils with and without fertilizers increased with time after inoculation. The increase was always greater for plants in soil with fertilizers. There was a correlation between the number of galls per gram of root and the reduction in plant growth due to the nematodes. Plants in fertilized soil which had more galls per root system but fewer galls per gram of root were less affected by the nematodes as was shown by the increases in shoot weight, root length and root weight. The fewer galls per gram of root therefore represents a dilution of intensity of attack per unit of root weight due to the effects of fertilizers on root growth. This shows that more galls per root system may not always reflect greater damage by the nematodes to plants. Assessments made for the damage to plants by root knot nematodes which are based on the number of galls per root system could be misleading. Such assessments may have been responsible for the non-recognition of fertilizer application as a means of alleviating the damage caused by root knot nematodes to plants. For example, Prasad *et al.* (1965), in evaluating the influence of nitrogen fertilizers on damage to tobacco by root knot nematodes based their assessments on the number of galls per root system.

The results which showed that plants with a greater percentage of roots not galled suffered less damage from the attack of the nematodes suggested that poor uptake and upward translocation of nutrients was involved in growth reduction. But Price (1979) found that the presence of *H. avenae* in roots of cereals did not impede upward nutrient translocation, indicating that growth reduction was due mainly to downward translocation of metabolites to females in syncytia acting as sinks. If this is the main cause of poor growth of plants infected with root knot nematodes, plants with more galls per root system should suffer more damage because of the greater number of metabolic sinks. But it was shown from this work that damage to eggplant was less on plants which had more galls per root system but fewer galls per gram of root suggesting that it is the distribution of the galls on the root system which is more important. It is my view therefore that two opposing forces operate when root knot nematodes attack plants. The downward translocation of metabolites to the sinks on one hand and the regeneration of new roots capable of utilizing nutrients in the soil on the other. The damage caused by nematodes to the plant is greater when the rate of downward translocation of metabolites is more than the rate of nutrient uptake by newly formed roots. Studies using radio-tracers should be done to elucidate the mode of nutrient translocation in plants infected with root-knot nematodes. Different population densities should be used as this would give a better understanding of how nutrients are translocated in infected plants at different levels of infestation in the field. The possibility of hormone involvement in stimulating growth of infected plants as suggested by Wallace (1971b) also needs further clarification.

A thorough appraisal of the results of the experiments on the interaction of fertilizers with *M. incognita* on vegetables indicate that NPK fertilizers applied at the recommended rates will help in minimizing crop losses due to the root knot nematodes on vegetables such as eggplant and possibly tomato under field conditions in Nigeria, particularly under rainfed conditions when soil populations of nematodes are low. The results of this work, however, show that agronomic practices by farmers in the tropics such as the application of nitrogen fertilizers 2 - 3 weeks after transplanting, poor nutrient balance in the soil as K is usually not applied when single fertilizers are used, are factors which reduce the effectiveness of fertilizers as a means of improving the tolerance of the plants to the damaging effects of the nematodes in tropical field conditions. The direct seeding of vegetable crops such as lettuce, carrot, Okra and cucumber, is another major factor contributing to the reduced effectiveness of fertilizers. The early application of fertilizers at planting and the use of nematode-free transplants where practicable should lead to higher yields of vegetables in soils infested with root-knot nematodes. Soil sampling to ensure that the correct proportion of nutrients are maintained in the soil will also enhance their effectiveness. The application of higher rates of fertilizers and increasing the amounts of nitrogen and potassium where economically feasible will further improve the tolerance of the plants to the nematodes and this will enable them to produce higher yields in spite of the presence of the nematodes.

The application of carbofuran at the rate of 8kg a.i/ha which falls within the range recommended by the manufacturers was found to be effective in increasing the growth of eggplant, lettuce and carrot infected with *M. incognita*, but was poor in reducing the nematode populations. The very high final soil population of juveniles found under eggplant after carbofuran application is in contrast with the findings of Averre *et al.* (1974); Reddy (1975); Reddy and Rao (1975) who found that final soil populations of root-knot nematodes were significantly reduced by carbofuran application. This may be due to differences in soil environmental conditions such as temperature, organic matter content of the soil, soil pH and also the rate and method of application. The poor control may also be due to the fact that carbofuran is not very effective against *M. incognita*. For example, Overman (1971) found that carbofuran was effective against *P. penetrans* for at least 21 weeks after treatment. Damadzadeh (1979) also found that carbofuran was less effective against *M. incognita* than against *Globodera rostochiensis* and *Ditylenchus dipsaci*. The varying responses of these nematodes to carbofuran treatment may be due to such factors as the permeability of the nematodes to the nematicide (Nelmes, 1971) and the rate of metabolism in the nematode (Le Patourel and Wright, 1976; Batterby *et al.*, 1977). Different species of *Meloidogyne* may respond differently to the treatment of the same nematicide as has been reported by McLeod and Khair (1975). The effect of carbofuran on different species of *Meloidogyne* should be compared as mixed species are commonly found in the same field.

High soil temperature and high organic matter content of the soil were found to be two factors which can reduce the efficacy of carbofuran. Thus carbofuran controlled *M. incognita* better at the soil temperature of 20°C than at 27° - 30°C, but nematode damage to eggplant was still significantly reduced and plant growth increased at the higher soil temperature regime. This is in agreement with the report of Damadzadah (1979) who found that carbofuran was more effective at preventing the invasion of tomato roots by *M. incognita* at low soil temperature. Miller and Rich (1974) also found that carbofuran was more effective against *P. penetrans* at low soil temperature. The reduced effectiveness at the higher soil temperatures may be related to faster degradation of carbofuran to non toxic metabolites.

The application of carbofuran was ineffective in controlling *M. incognita* in soil containing 50% organic matter but significantly increased growth of eggplant in soil with 25% organic matter. The importance of organic matter content of the soil as a factor which can limit the efficacy of carbofuran has not been previously reported. Whitehead *et al.* (1973b) also found that the efficacy of phenamiphos, an organophosphorus nematicide against *Globodera rostochiensis* was reduced in peaty loam soil. The efficacy of phenamiphos against *D. dipsaci* was similarly reduced in soil with high organic matter content (Bunt, 1975). The poorer control of *M. incognita* by carbofuran in soil with high organic matter indicated that a high proportion of the nematicide was absorbed onto the organic matter fraction of the soil. This result also showed the carbofuran has a high partition coefficient (Q value) which according to Bromilow (1973) influences the effectiveness of pesticides in soil with different organic matter content.

It was shown that carbofuran applied at field rates controls *M. incognita* in the soil and has no effect on nematodes already in the roots. This is in agreement with the views held by other workers (Whitehead, 1973; Hague and Pain, 1973; Bromilow and Lord, 1979) who consider that organophosphate and carbamate nematicides affect nematodes principally in the soil. As the results showed that the metabolite 3-hydroxycarbofuran plays no role in the control of *M. incognita* at field rates, conditions favouring its rapid formation such as high soil pH (Caro *et al.*, 1973) will reduce the efficacy of carbofuran. The role of 3-ketocarbofuran which is another major metabolite of carbofuran in the control of nematodes should be ascertained.

Egg hatching of *M. incognita* was delayed at a low concentration of carbofuran indicating that this is one of the ways it reduces damage caused by these nematodes to crop plants. Since egg hatching and root invasion by second stage juveniles were prevented at concentrations of the nematicide which did not affect mobility, the effect of carbofuran on the behavioural sequences necessary for hatching and invasion is therefore its principal mode of action at field rates. Second stage juveniles of *M. incognita* placed on agar plates containing carbofuran at concentrations of 1 - 8ppm migrated away from the roots of tomato seedlings and their movement was non-directional. On agar plates without carbofuran, all the nematodes remained near the roots and up to 58% of them invaded the roots during the experimental period. These behavioural responses of *M. incognita* to carbofuran on agar plates indicated that it was the impairment of the sensory components which was involved in preventing root invasion at low concentrations. Di Sanzo (1973), from his work on the behavioural responses of *P. penetrans* and *T. claytoni* to carbofuran also suggested that the impairment of the sensory components of the nematodes might be involved in the action of the nematicide. A similar suggestion has been made for the effects of low concentrations of oxamyl on root invasion by *M. incognita* (Wright *et al.*, 1980).

From the experiments on the interaction between fertilizers and carbofuran on eggplant, lettuce and carrots, it was shown that they acted independently of one another in the way they influenced the damage caused by *M. incognita* to the crops. The results in Table 3.2.1 showed more females in the roots of plants given the combined application of fertilizers and carbofuran than in the roots of plants in soil treated with only carbofuran. The application of fertilizers may have enhanced the uptake of the nematicide into the roots and its translocation to the leaves. Further investigations are needed to elucidate this.

The combined effect of fertilizers and carbofuran on damage caused by *M. incognita* to eggplant and lettuce was additive. The reduction in nematode damage following their combined application was greater on eggplant than lettuce, while on carrot the reduction in damage was due mainly to carbofuran action. The observed difference between the effects of carbofuran and fertilizers on the three crops can be related to their method of cultivation i.e. transplanted seedlings and direct seeding. The slow development of carrot during the early stages of growth may have been responsible for the observed effects. The role of fertilizers may have been to enhance the growth of eggplant and lettuce so that the vulnerable stages were quickly passed. The period of activity of the nematodes after recovery was therefore synchronized with the stage of the plants growth when it was fully established and less susceptible to damage. It is suggested that quick-growing varieties of vegetables and where practicable, transplants be used with carbofuran as this might lead to less crop losses that can result from the damaging effects of nematodes upon recovery from the nematicide. The results also emphasize the importance of using adequate levels of fertilizers to promote rapid growth of plants so that their vulnerable stages can be quickly passed.

From the above results it can be seen that carbofuran can be used quite effectively for reducing the damage caused by *M. incognita* to some vegetables in the tropics. High final soil populations are likely to be left in the soil because of the high soil temperatures prevailing in the tropics which will reduce the stability of the nematicide and also enhance nematode reproduction. Since carbofuran at low concentrations can delay egg hatching, its use should be directed at delaying hatching rather than preventing root invasion of already hatched second stage juveniles. This will prolong the time between planting and invasion resulting in less damage to the crop. In Nigeria, where root-knot nematodes survive the dry season in the egg stage, the application of carbofuran before the first rains will result in better control of the nematodes.

The practice of applying large amounts of organic manures such as cow dung and poultry droppings is common among certain farmers in Nigeria and may need to be discouraged in intergrated control programmes for root-knot nematodes as their application could greatly reduce the efficacy of the nematicides, particularly carbofuran. Quick growing varieties of vegetables and where practicable, transplants should be used in soils treated with carbofuran to minimize losses that can result from the effects of the nematodes upon their recovery from the nematicide.

The usefulness of a nematicide for controlling nematodes is usually based on its ability to increase crop yields profitably and keep down soil populations of the nematodes. The results of this work and those of other workers (Averre *et al.*, 1974; Reddy and Rao, 1975) indicate that carbofuran is less effective than aldicarb and oxamyl, particularly in reducing soil populations. But aldicarb and oxamyl have high oral and dermal toxicities and would be unsuitable for use in countries where the bulk of the workforce are not literate. Despite the lower efficacy of carbofuran in controlling root-knot nematodes, I would recommend its use in Nigeria and other African countries because of the low dermal toxicity. However, the problem of residues which might be left in the crop should not be overlooked, particularly on certain crops e.g. lettuce and carrot, which can be eaten raw, as carbofuran has a high oral toxicity. Particular caution must be exercised when multiple application is used.

Although the application of fertilizers and carbofuran have been found to be effective in increasing crop yields, their use must be shown to be profitable before they are included in a management programme for reducing crop losses due to root-knot nematodes in Nigeria. The decision whether to apply fertilizers alone or in combination with nematicides will be based on economical considerations and the possible impact each method will have on the farming community. The application of fertilizers should be profitable for reducing the damage caused by root-knot nematodes to vegetables in Nigeria as fertilizers are at present cheap because of high Government subsidy. From the data in Table 3.2.2, eggplant given fertilizers at transplanting will give a yield of 3423.5kg/ha (41,000 plants/ha). Based on the current prices of eggplant and fertilizers in Nigeria, it would cost N90 to apply fertilizers at the rate used in this work while a revenue of N342.25 will be realized. The application of carbofuran should also be profitable for use on high valued cash crops like tomato, cowpea and broad beans, particularly as further application of insecticides may be eliminated or minimized. Field experiments are needed in Nigeria to work out the economics of using fertilizers and carbofuran for the production of different vegetables in soils infested with root-knot nematodes.

As a result of the investigations described in this thesis, the basic management programme I would recommend for the control of root-knot nematodes on vegetables in Nigeria is one which combines the use of fertilizers, resistant varieties and a nematicide, specifically carbofuran. The resistant varieties of tomato which can be grown successfully in Nigeria have been identified but it would be unwise to recommend their frequent cultivation. Fertilizers should be used for the rainfed crop or after the cultivation of suitable resistant tomato varieties when soil populations of the nematodes will be low. Ideally, this should be in irrigated fields in the north of Nigeria and areas with lakes and streams where rainfall can be supplemented by irrigation if necessary. Inadequate moisture or heavy rainfall as found in the south of Nigeria will reduce the availability of the nutrients to the plants. Carbofuran should be used when soil populations of the nematodes are very high such as after a susceptible crop.

The results of this work have helped to define clearly the role of fertilizers as applied in the production of vegetables in tropical soils infested with *M. incognita*. Their role in the control of *M. incognita* on vegetables with carbofuran has also been highlighted. The basis for the tolerance of plants to the damaging effects of root-knot nematodes when fertilizers are applied has been elucidated. The correlation between the percentage of roots not galled, the number of galls per gram of root and damage to the plant could be useful in screening crop plants for tolerance to root-knot nematode infection.

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APPENDIX A

TABLE 1 INFLUENCE OF TIME OF APPLICATION OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF EGGPLANT INFECTED WITH M.INCOGNITA

FRESH WEIGHT OF TOPS AT HARVESTING (STEM, LEAVES AND FRUITS)

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizers only at transplanting	147.1	123.4	162.9	137.2	142.7
2. Fertilizers only 2 weeks after transplanting	133.4	104.0	121.9	141.6	125.2
3. Nematicide only at transplanting	77.9	96.4	84.2	90.3	87.2
4. Nematicide 2 weeks after transplanting	75.2	69.4	54.9	78.8	69.6
5. Fertilizer and Nematicide applied together at transplanting	233.8	219.6	199.7	241.7	223.7
6. Fertilizer at transplanting plus Nematicides 2 weeks later	151.2	191.6	204.9	179.9	181.9
7. Nematicide at transplanting plus fertilizer 2 weeks later	257.6	211.7	249.2	238.7	239.3
8. None	34.6	46.3	31.7	36.2	37.2

APPENDIX A

TABLE 2 INFLUENCE OF TIME OF APPLICATION OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF EGGPLANT INFECTED WITH M.INCOGNITA

FRESH WEIGHT OF ROOTS

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizers only at transplanting	54.5	47.6	58.9	42.7	50.93
2. Fertilizers only 2 weeks after transplanting	49.6	51.6	41.8	53.2	49.05
3. Nematicide only at transplanting	39.8	41.7	36.2	37.1	38.70
4. Nematicide 2 weeks after transplanting	29.2	31.7	28.6	24.7	28.55
5. Fertilizer and Nematicide applied together at transplanting	49.2	51.8	52.6	54.6	52.10
6. Fertilizer at transplanting plus Nematicide 2 weeks later	47.2	51.9	48.6	51.7	49.90
7. Nematicide at transplanting plus Fertilizer 2 weeks later	48.8	46.2	56.8	50.2	50.50
8. None	24.6	21.6	20.8	22.7	22.43

APPENDIX A

TABLE 3 INFLUENCE OF THE TIME OF APPLICATION OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF EGGPLANT INFECTED WITH M. INCOGNITA

FINAL SOIL POPULATION OF JUVENILES PER 200g OF SOIL

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizers only at transplanting	2985	1900	2600	2090	2394
2. Fertilizers only 2 weeks after transplanting	2850	1150	1290	2690	2060
3. Nematicide only at transplanting	840	1295	1200	955	1070
4. Nematicide 2 weeks after transplanting	1020	900	1325	1210	1114
5. Fertilizer and Nematicide applied together at transplanting	2650	1755	1350	2450	2051
6. Fertilizer at transplanting plus Nematicide 2 weeks later	2850	2050	1990	3025	2479
7. Nematicide at transplanting plus Fertilizer 2 weeks later	1795	3250	2785	2900	2693
8. None	1185	600	1295	1015	1024

APPENDIX A

TABLE 4 INFLUENCE OF TIME OF APPLICATION OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF EGGPLANT INFECTED WITH M.INCOGNITA

RATINGS FOR ROOT GALLING

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizers only at transplanting	7.0	7.0	8.0	7.0	7.25
2. Fertilizers only 2 weeks after transplanting	7.0	7.0	8.0	8.0	7.50
3. Nematicide only at transplanting	7.0	7.0	7.0	8.0	7.25
4. Nematicide 2 weeks after transplanting	8.0	7.0	7.0	8.0	7.50
5. Fertilizer and Nematicide applied together at transplanting	7.0	7.0	7.0	7.0	7.00
6. Fertilizer at transplanting plus Nematicide 2 weeks later	8.0	7.0	7.0	7.0	7.25
7. Nematicides at transplanting plus Fertilizer 2 weeks later	7.0	7.0	7.0	7.0	7.00
8. None	8.0	9.0	8.0	8.0	8.25

APPENDIX A

TABLE 5 INFLUENCE OF TIME OF APPLICATION OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF EGGPLANT INFECTED WITH M. INCOGNITA

MARKETABLE YIELDS AT HARVESTING

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizers only at transplanting	91.2	78.9	60.9	102.9	83.5
2. Fertilizers only 2 weeks after transplanting	68.7	59.8	58.5	73.2	65.0
3. Nematicide only at transplanting	0	0	0	0	0
4. Nematicide 2 weeks after transplanting	0	0	0	0	0
5. Fertilizer and Nematicide applied together at transplanting	148.3	132.1	122.9	156.3	139.9
6. Fertilizer at transplanting plus Nematicide 2 weeks later	96.9	111.9	119.5	104.7	108.3
7. Nematicide at transplanting plus Fertilizer 2 weeks later	178.5	131.9	162.8	126.6	150.0
8. None	0	0	0	0	0

APPENDIX B

TABLE 1 EFFECTS OF FERTILIZERS AND CARBOFURAN APPLIED SINGLY AND IN COMBINATION ON THE GROWTH OF EGGPLANT INFECTED WITH *M. INCOGNITA*

FRESH WEIGHT OF TOPS

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	64.6	86.2	71.8	74.2	74.2
2. Fertilizer - Nematodes	82.2	112.8	94.8	97.0	96.7
3. Carbofuran + Nematodes	37.3	42.9	51.2	39.8	42.8
4. Carbofuran - Nematodes	65.4	60.9	72.8	59.7	64.7
5. Fertilizer + Carbofuran + Nematodes	99.3	89.2	104.8	77.9	92.8
6. Fertilizer + Carbofuran - Nematodes	108.0	99.8	121.7	89.7	104.8
7. None + Nematodes	14.9	21.2	19.2	20.3	18.9
8. None - Nematodes	49.5	69.4	72.2	59.3	62.6

TABLE 2 EFFECTS OF FERTILIZERS AND CARBOFURAN APPLIED SINGLY AND IN COMBINATION ON THE GROWTH OF EGGPLANT INFECTED WITH M. INCOGNITA

FRESH WEIGHT OF ROOT

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	44.2	38.9	39.1	43.4	41.4
2. Fertilizer - Nematodes	35.8	33.7	30.9	31.2	32.9
3. Carbofuran + Nematodes	23.4	19.6	17.2	24.6	21.2
4. Carbofuran - Nematodes	16.9	22.8	17.2	22.8	19.9
5. Fertilizer + Carbofuran + Nematodes	42.6	36.1	35.9	38.7	38.2
6. Fertilizer + Carbofuran - Nematodes	35.8	30.9	34.9	32.0	33.2
7. None + Nematodes	11.4	12.9	13.6	10.9	12.2
8. None - Nematodes	14.0	15.8	13.9	15.6	17.7

APPENDIX B

TABLE 4 EFFECTS OF FERTILIZERS AND CARBOFURAN APPLIED SINGLY AND IN COMBINATION ON THE GROWTH OF EGGPLANT INFECTED WITH M.INCOGNITA

RATINGS FOR ROOT GALLING

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	5.0	6.0	5.0	5.0	5.2
2. Carbofuran + Nematodes	4.0	4.0	4.0	4.0	4.0
3. Fertilizer + Carbofuran + Nematodes	4.0	4.0	4.0	4.0	4.0
4. None + Nematodes	5.0	6.0	6.0	5.0	5.4

APPENDIX C

TABLE 1 EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF LETTUCE INFECTED WITH M.INCOGNITA

FRESH WEIGHT OF LEAVES

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	19.1	27.2	19.9	23.4	22.40
2. Fertilizer - Nematodes	30.7	39.6	47.2	32.9	37.60
3. Carbofuran + Nematodes	18.4	22.2	17.2	20.8	19.65
4. Carbofuran - Nematodes	25.8	28.2	21.9	25.3	25.30
5. Fertilizer + Carbofuran + Nematodes	38.9	32.4	35.9	29.6	34.20
6. Fertilizer + Carbofuran - Nematodes	37.5	40.1	39.3	36.9	38.45
7. None + Nematodes	8.9	10.2	6.0	7.3	8.10
8. None - Nematodes	22.5	25.3	28.4	22.7	24.73

TABLE 2 EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF LETTUCE INFECTED WITH *M. INCOGNITA*

FRESH ROOT WEIGHT

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	9.2	9.9	11.2	8.6	9.60
2. Fertilizer - Nematodes	7.9	8.6	8.9	8.2	8.40
3. Carbofuran + Nematodes	6.7	7.1	7.9	5.9	6.90
4. Carbofuran - Nematodes	6.3	5.6	6.8	5.7	6.10
5. Fertilizer + Carbofuran + Nematodes	8.6	11.2	9.7	10.9	10.10
6. Fertilizer + Carbofuran - Nematodes	9.7	10.0	8.9	8.2	9.20
7. None + Nematodes	7.1	6.4	6.2	7.5	6.80
8. None - Nematodes	6.5	5.8	6.2	5.8	6.07

TABLE 5 EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF LETTUCE INFECTED WITH M.INCOGNITA

FINAL SOIL POPULATION OF L_2 JUVENILES/200g OF SOIL

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	330	295	350	385	340.0
2. Carbofuran + Nematodes	80	200	165	95	135.0
3. Fertilizer + Carbofuran + Nematodes	220	135	95	160	152.0
4. None + Nematodes	165	250	110	185	177.5

APPENDIX C

TABLE 6 EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF LETTUCE INFECTED WITH *M. INCOGNITA*

RATINGS FOR ROOT GALLING

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	6.0	6.0	6.0	6.0	6.0
2. Carbofuran + Nematodes	5.0	5.0	5.0	5.0	5.0
3. Fertilizer + Carbofuran + Nematodes	5.0	5.0	5.0	6.0	5.5
4. None + Nematodes	6.0	6.0	6.0	6.0	6.0

APPENDIX C

TABLE 7 EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF CARROT INFECTED WITH *M. INCOGNITA*

FRESH WEIGHT OF STORAGE ROOTS

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	9.6	7.3	10.4	7.9	8.8
2. Fertilizer - Nematodes	16.9	12.9	14.2	17.2	15.3
3. Carbofuran + Nematodes	5.4	6.7	5.9	7.2	6.3
4. Carbofuran - Nematodes	9.9	12.2	8.7	10.8	10.4
5. Fertilizer + Carbofuran + Nematodes	13.8	11.9	12.2	15.3	13.3
6. Fertilizer + Carbofuran - Nematodes	15.5	16.9	17.2	19.9	17.2
7. None + Nematodes	2.7	1.4	2.3	2.0	2.1
8. None - Nematodes	10.6	8.6	11.2	9.2	9.9

APPENDIX C

TABLE 8 EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF CARROT INFECTED WITH *M. INCOGNITA*

FRESH WEIGHT OF LEAVES

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizers + Nematodes	6.4	5.0	3.9	7.5	5.7
2. Fertilizers - Nematodes	8.4	7.9	9.4	5.9	7.9
3. Carbofuran + Nematodes	2.1	2.6	1.6	2.1	2.1
4. Carbofuran - Nematodes	2.7	2.4	3.4	3.1	2.9
5. Fertilizer + Carbofuran + Nematodes	6.3	7.4	5.9	5.2	6.2
6. Fertilizer + Carbofuran - Nematodes	8.5	5.7	8.1	6.1	7.1
7. None + Nematodes	2.5	1.4	1.7	2.0	1.9
8. None - Nematodes	4.3	3.8	4.6	2.9	3.9

APPENDIX C

TABLE 9 EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF CARROT INFECTED WITH <u>M.INCOGNITA</u>

LENGTH OF STORAGE ROOT IN MM.

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	22.5	21.9	19.2	22.0	24.4
2. Fertilizer - Nematodes	98.2	101.2	92.9	84.6	94.2
3. Carbofuran + Nematodes	31.3	37.2	29.3	34.2	33.0
4. Carbofuran - Nematodes	56.2	40.9	61.2	58.3	54.1
5. Fertilizer + Carbofuran + Nematodes	72.1	66.7	78.2	70.4	71.8
6. Fertilizer + Carbofuran - Nematodes	104.4	87.9	91.9	99.8	96.0
7. None + Nematodes	21.2	14.9	18.9	20.9	18.9
8. None - Nematodes	42.8	51.6	58.9	49.6	50.7

APPENDIX C

TABLE 10 EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF CARROT INFECTED WITH *M. INCOGNITA*

FINAL SOIL POPULATION OF L_2 JUVENILES/200g OF SOIL

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	185	225	140	115	166.3
2. Carbofuran + Nematodes	70	55	75	55	71.3
3. Fertilizer + Carbofuran + Nematodes	95	105	85	155	85.0
4. None + Nematodes	85	100	70	135	95.5

APPENDIX C

TABLE 11 EFFECTS OF FERTILIZERS AND CARBOFURAN ON THE GROWTH OF CARROT INFECTED WITH *M. INCOGNITA*

SCORES FOR ROOT DEFORMATION

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
1. Fertilizer + Nematodes	3.0	3.0	3.0	3.0	3.0
2. Carbofuran + Nematodes	2.0	2.0	2.0	2.0	2.0
3. Fertilizer + Carbofuran + Nematodes	2.0	2.0	2.0	2.0	2.0
4. None + Nematodes	3.0	3.0	3.0	3.0	3.0

APPENDIX D

TABLE 1 MECHANISM OF ACTION OF FERTILIZERS ON *M. INCOGNITA*

EFFECTS OF FERTILIZERS ON ROOT INVASION

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
No Fertilizer applied	200	226	210	182	204.5
Normal rate (18.29g/pot)	196	176	210	190	193
1.5 x normal rate	183	190	200	173	186.5
2 x normal rate	156	163	186	170	168.8

APPENDIX D

TABLE 2 MECHANISM OF ACTION OF FERTILIZERS ON M.INCOGNITA

INCREASE IN FRESH SHOOT WEIGHT WITH TIME

TREATMENTS	REPLICATES	3 Weeks	6 Weeks
Fertilizer + Nematodes	1	25.6	58.2
	2	31.2	64.0
	3	20.1	53.2
	4	28.7	63.4
	Mean	26.4	59.7
Fertilizer - Nematodes	1	26.9	78.9
	2	30.4	70.7
	3	35.0	76.1
	4	28.9	73.7
	Mean	30.3	74.8
None + Nematodes	1	11.9	18.5
	2	15.2	16.9
	3	17.8	23.2
	4	10.7	14.2
	Mean	13.9	18.2
None - Nematodes	1	20.8	46.4
	2	14.9	51.3
	3	17.5	42.9
	4	23.2	56.2
	Mean	19.1	49.2

APPENDIX D

TABLE 3 MECHANISM OF ACTION OF FERTILIZERS ON M.INCOGNITA

INCREASE IN DRY SHOOT WEIGHT WITH TIME

TREATMENTS	REPLICATES	3 Weeks	6 Weeks
Fertilizer + Nematodes	1	4.31	9.49
	2	5.13	10.87
	3	3.44	8.91
	4	4.69	10.61
	Mean	4.39	9.97
Fertilizer - Nematodes	1	4.50	13.26
	2	5.03	11.83
	3	5.78	12.71
	4	4.39	12.33
	Mean	4.92	12.65
None + Nematodes	1	2.01	3.10
	2	2.61	2.91
	3	3.11	3.76
	4	1.72	2.39
	Mean	2.36	3.02
None - Nematodes	1	3.39	7.81
	2	2.59	8.49
	3	2.84	7.21
	4	3.69	9.42
	Mean	3.12	8.23

APPENDIX D

TABLE 4 MECHANISM OF ACTION OF FERTILIZERS ON *M. INCOGNITA*

INCREASE IN FRESH ROOT WEIGHT WITH TIME

TREATMENTS	REPLICATES	3 Weeks	6 Weeks	9 Weeks
Fertilizer + Nematodes	1	11.4	26.9	48.4
	2	13.8	21.5	39.9
	3	10.8	30.2	36.8
	4	15.2	28.2	47.1
	Mean	12.8	26.7	43.0
Fertilizer - Nematodes	1	9.8	18.6	29.6
	2	11.2	20.8	36.9
	3	10.4	18.5	30.8
	4	9.0	24.2	32.2
	Mean	10.1	20.5	32.4
None + Nematodes	1	7.4	15.2	14.9
	2	5.9	10.7	22.9
	3	6.6	12.9	20.6
	4	5.9	14.0	17.6
	Mean	6.4	13.2	18.9
None - Nematodes	1	6.2	11.6	16.2
	2	5.3	10.9	17.3
	3	4.9	13.8	18.2
	4	6.4	10.1	13.9
	Mean	5.7	11.6	16.4

APPENDIX D

TABLE 5 MECHANISM OF ACTION OF FERTILIZERS ON *M. INCOGNITA*

INCREASE IN ROOT LENGTH WITH TIME

TREATMENTS	REPLICATES	3 Weeks	6 Weeks	9 Weeks
Fertilizer + Nematodes	1	18.9	24.9	30.8
	2	20.7	29.8	38.2
	3	18.6	26.8	31.2
	4	23.4	30.1	32.6
	Mean	20.4	27.9	33.2
Fertilizer - Nematodes	1	22.8	36.9	36.2
	2	20.9	27.6	40.5
	3	24.9	33.2	42.8
	4	22.6	31.9	38.9
	Mean	22.8	32.4	39.6
None + Nematodes	1	15.5	10.6	15.9
	2	9.9	18.0	12.2
	3	13.2	12.1	18.8
	4	12.6	14.9	10.7
	Mean	12.8	13.9	14.4
None - Nematodes	1	18.9	24.8	26.9
	2	14.2	16.9	32.9
	3	21.8	28.2	23.7
	4	12.7	19.7	34.9
	Mean	16.9	22.4	29.6

APPENDIX D

TABLE 6 MECHANISM OF ACTION OF FERTILIZERS ON *M.INCOGNITA*

INCREASE IN NUMBER OF GALLS PER ROOT SYSTEM WITH TIME

TREATMENTS	REPLICATES	3 Weeks	6 Weeks
Fertilizer + Nematodes	1	592	2313
	2	621	1892
	3	604	2083
	4	653	2088
	Mean	617	2086
None + Nematodes	1	584	1763
	2	619	1498
	3	567	1625
	4	554	1652
	Mean	581	1634

INCREASE IN NUMBER OF GALLS PER GRAM OF ROOT WITH TIME

TREATMENTS	REPLICATES	3 Weeks	6 Weeks
Fertilizer + Nematodes	1	52	86
	2	45	88
	3	56	69
	4	43	73
	Mean	49	79
None + Nematodes	1	79	116
	2	105	140
	3	86	126
	4	94	118
	Mean	91	125

APPENDIX D

TABLE 7 MECHANISM OF ACTION OF FERTILIZERS ON M.INCOGNITA

INCREASE IN PROPORTION OF ROOTS GALLED WITH TIME (SCORES)

TREATMENTS	REPLICATES	3 Weeks	6 Weeks	9 Weeks
Fertilizer + Nematodes	1	1.0	2.0	4.0
	2	1.0	3.0	4.0
	3	1.0	2.0	4.0
	4	1.0	3.0	4.0
	Mean	1.0	2.5	4.0
None + Nematodes	1	2.0	4.0	5.0
	2	2.0	4.0	5.0
	3	2.0	4.0	5.0
	4	1.0	3.0	5.0
	Mean	1.75	3.75	5.0

APPENDIX D

TABLE 8 MECHANISM OF ACTION OF FERTILIZERS ON <u>M. INCOGNITA</u>

IN VITRO EFFECTS OF FERTILIZERS ON <u>M. INCOGNITA</u> JUVENILES - NO. OF ACTIVE NEMATODES

		CONCENTRATIONS OF FERTILIZER				
TIME	REPLI-CATES	0	10 ppm	100 ppm	1000 ppm	10,000 ppm
6	1	10	10	10	10	10
	2	10	10	10	10	10
	3	10	10	10	10	10
24	1	10	10	10	6	3
	2	10	10	10	4	5
	3	10	10	10	5	3
48	1	10	10	10	5	2
	2	10	10	10	4	3
	3	10	10	10	3	2
72	1	10	10	8	3	2
	2	10	10	7	3	1
	3	10	10	8	3	4
96	1	10	10	7	2	0
	2	10	10	7	3	0
	3	10	10	8	2	0
7 day	1	7	9	7	2	0
	2	8	8	6	1	0
	3	8	7	6	2	0

APPENDIX E

TABLE 1 MECHANISM OF ACTION OF CARBOFURAN

EFFECT OF CARBOFURAN AND 3 HYDROXYCARBOFURAN ON ROOT INVASION BY M.INCOGNITA

	TREATMENTS	REPLICATES				MEAN
		1	2	3	4	
CARBOFURAN	Control I 1% Phostrogen	65	60	55	79	64.7
	Control II 1% Acetone	56	63	68	58	61.2
	2ug/g of sand	15	19	22	20	19.0
	4ug/g of sand	11	14	9	11	11.2
	8ug/g of sand	3	4	6	7	5.0
3 HYDROXY-CARBOFURAN	2ug/g of sand	53	69	55	68	61.2
	4ug/g of sand	62	61	55	68	61.5
	8ug/g of sand	55	67	59	47	57.0

APPENDIX E

TABLE 2 MECHANISM OF ACTION OF CARBOFURAN

INFLUENCE OF CONCENTRATION AND EXPOSURE TIME ON INVASION

1 DAY EXPOSURE TIME

CONCENTRATION OF CARBOFURAN	REPLICATES			MEAN
	1	2	3	
Control	67	56	62	61.6
2 ppm	76	68	57	67.0
4 ppm	57	68	53	59.3
8 ppm	59	50	62	57.0

7 DAY EXPOSURE TIME

CONCENTRATION OF CARBOFURAN	REPLICATES			MEAN
	1	2	3	
0	59	48	53	53.3
2 ppm	37	41	48	42.0
4 ppm	38	48	50	45.3
8 ppm	46	39	39	41.3

APPENDIX E

TABLE 3 MECHANISM OF ACTION OF CARBOFURAN

BARE ROOT DIPPING TEST

2 DAYS AFTER REPLANTING

TREATMENTS	REPLICATES			MEAN
	1	2	3	
Control	375	321	404	366.6
2 ppm	330	420	326	358.6
4 ppm	350	236	227	271.0
8 ppm	250	334	329	304.3

9 DAYS AFTER REPLANTING

TREATMENTS	REPLICATES			MEAN
	1	2	3	
Control	294	382	297	324.3
2 ppm	328	385	306	339.6
4 ppm	254	297	332	294.3
8 ppm	296	218	329	281.0

APPENDIX E

TABLE 4 MECHANISM OF ACTION OF CARBOFURAN

MIGRATION OF SECOND STAGE JUVENILES THROUGH A SAND COLUMN

TREATMENTS	REPLICATES			MEAN
	1	2	3	
0 ppm	37	34	32	34.3
1 ppm	35	35	38	36.0
2 ppm	29	34	32	31.6
4 ppm	24	19	20	21.0
8 ppm	14	16	10	13.3

APPENDIX E

TABLE 5 EFFECT OF CARBOFURAN ON HATCHING

(5a) IN PESTICIDE - NO. OF JUVENILES

TREATMENTS	REPLICATES			MEAN
	1	2	3	
0 (Control)	586	499	663	582.6
Carbofuran at 1 ppm	624	478	605	569.0
" " 2 ppm	484	229	342	375.0
" " 4 ppm	338	265	340	314.3
" " 8 ppm	166	126	202	164.6

(5b) IN WATER - NO. OF JUVENILES

TREATMENTS	REPLICATES			MEAN
	1	2	3	
0 (Control)	345	524	409	396.6
Carbofuran at 1 ppm	489	350	604	481.0
" " 2 ppm	535	703	596	611.3
" " 4 ppm	772	845	696	771.0
" " 8 ppm	882	652	952	828.6

(5c) TOTAL HATCH - NO. OF JUVENILES

TREATMENTS	REPLICATES		
	1	2	3
0 (Control)	582.6	392.6	975.2
Carbofuran at 1 ppm	569.0	481.0	1050.0
" " 2 ppm	378.3	611.3	989.6
" " 4 ppm	314.3	771.0	1085.3
" " 8 ppm	164.6	828.6	993.2

APPENDIX E

MECHANISM OF ACTION OF CARBOFURAN

SYSTEMIC EFFECT OF CARBOFURAN ON *M. INCOGNITA*

TABLE 1 THREE DAYS ALLOWED FOR INVASION (NO. OF JUVENILES AND FEMALES PER ROOT SYSTEM)

TREATMENTS	Rate of Carbofuran kg a.i/ha	No. of Females	No. of J_2-J_4 Juveniles
1. Plants from untreated into treated soil	4	49, 48, 41	24, 19, 23
2. "	8	43, 53, 45	27, 23, 34
3. "	16	51, 42, 47	26, 18, 25
4. Plants from untreated into untreated soil	0	43, 53, 39	30, 21, 26

TABLE 2 SEVEN DAYS ALLOWED FOR INVASION (NO. OF JUVENILES AND FEMALES PER ROOT SYSTEM)

TREATMENTS	Rate of Carbofuran kg a.i/ha^{-1}	No. of Females	No. of J_2-J_4 Juveniles
1. Plants from untreated into treated soil	4	66, 70, 63	26, 23, 26
2. "	8	80, 76, 76	30, 36, 33
3. "	16	86, 83, 90	33, 40, 30
4. Plants from untreated into untreated soil	0	83, 76, 86	20, 26, 33

APPENDIX F

FACTORS INFLUENCING THE EFFECTS OF FERTILIZERS ON *M. INCOGNITA*

TABLE 1 EFFECTS OF FERTILIZER RATES

FRESH WEIGHT OF TOPS

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	97.9	81.6	79.3	94.8
2 Fertilizers - Nematodes	91.8	118.7	102.6	94.1
2 Fertilizers + Nematodes	77.9	86.9	79.6	97.2
2 Fertilizers - Nematodes	96.2	101.6	112.7	91.9
Fertilizer + Nematodes	76.1	61.8	66.8	72.9
Fertilizer - Nematodes	99.6	84.9	102.6	95.3
Fertilizer + Carbofuran + Nematodes	104.2	86.9	97.7	81.6
Fertilizer + Carbofuran - Nematodes	101.5	110.8	103.6	96.9
None + Nematodes	10.9	6.4	8.1	13.0
None - Nematodes	64.8	50.9	63.7	54.2

TABLE 2 NO OF DEVELOPING FRUITS

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
2 Fertilizers + Nematodes	3.0	2.0	3.0	3.0	2.8
2 Fertilizers - Nematodes	4.0	5.0	5.0	3.0	4.2
2 Fertilizers + Nematodes	4.0	4.0	3.0	5.0	4.0
2 Fertilizers - Nematodes	5.0	4.0	3.0	4.0	4.0
Fertilizer + Nematodes	2.0	1.0	2.0	1.0	1.5
Fertilizer - Nematodes	3.0	2.0	3.0	4.0	3.0
Fertilizer + Carbofuran + Nematodes	4.0	5.0	4.0	4.0	4.2
Fertilizer + Carbofuran - Nematodes	6.0	4.0	3.0	6.0	4.7
None + Nematodes	0	0	0	0	0
None - Nematodes	0	0	1	1	0.5

APPENDIX F

TABLE 3 FRESH ROOT WEIGHTS

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	53.2	46.8	45.9	56.5
2 Fertilizers - Nematodes	39.7	43.1	38.9	39.1
2 Fertilizers + Nematodes	59.6	66.9	62.7	57.2
2 Fertilizers - Nematodes	56.2	46.2	49.9	48.1
Fertilizer + Nematodes	46.2	38.2	40.7	45.2
Fertilizer - Nematodes	30.6	39.8	30.7	31.9
Fertilizer + Carbofuran - Nematodes	41.1	39.6	32.8	31.9
Fertilizer + Carbofuran - Nematodes	38.2	30.6	28.6	36.2
None + Nematodes	8.4	7.8	6.8	9.2
None - Nematodes	13.8	11.9	14.1	15.8

APPENDIX F

TABLE 4 RATINGS FOR ROOT GALLING AND % OF ROOTS GALLED

TREATMENTS	ROOT GALLING REPLICATES				% OF ROOTS GALLED REPLICATES			
	1	2	3	4	1	2	3	4
2 Fertilizers + Nematodes	5.0	5.0	5.0	5.0	3.0	3.0	3.0	3.0
2 Fertilizers + Nematodes	5.0	5.0	5.0	5.0	3.0	3.0	3.0	3.0
Fertilizer + Nematodes	5.0	5.0	5.0	5.0	4.0	4.0	4.0	4.0
Fertilizer + Carbofuran + Nematodes	4.0	4.0	4.0	4.0	3.0	3.0	3.0	3.0
None + Nematodes	6.0	6.0	6.0	6.0	5.0	5.0	5.0	5.0

TABLE 5 FINAL SOIL POPULATIONS OF JUVENILES PER 200g OF SOIL

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	2650	1500	2455	1595
2 Fertilizers + Nematodes	3100	2050	2255	2195
Fertilizer + Nematodes	1895	1150	1925	1630
Fertilizer + Carbofuran + Nematodes	1855	1600	2150	1645
None + Nematodes	595	390	725	450

APPENDIX G1

HIGH SOIL POPULATION DENSITY

TABLE 1 FRESH WEIGHT OF TOPS

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	71.3	62.9	58.6	68.4
2 Fertilizers - Nematodes	115.7	92.7	102.4	119.6
Fertilizer + Nematodes	52.7	41.8	51.2	48.6
Fertilizer - Nematodes	93.4	84.2	99.2	89.6
None + Nematodes	0.0	0.0	0.0	0.0
None - Nematodes	49.3	58.6	43.1	50.6

TABLE 2 RATINGS FOR ROOT GALLING AND % OF ROOTS GALLED

TREATMENTS	ROOT GALLING REPLICATES				% OF ROOTS GALLED REPLICATES			
	1	2	3	4	1	2	3	4
2 Fertilizers + Nematodes	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Fertilizer + Nematodes	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
None + Nematodes	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

TABLE 3 FINAL SOIL POPULATIONS OF L_2 JUVENILES/200g OF SOIL

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	3270	2460	4320	3950
Fertilizer + Nematodes	2695	2955	3120	3620
None + Nematodes	0	0	0	0

TABLE 4 FRESH ROOT WEIGHTS

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	43.7	49.9	46.1	45.9
2 Fertilizers - Nematodes	40.6	43.1	32.9	37.0
Fertilizer + Nematodes	39.6	29.8	36.2	28.8
Fertilizer - Nematodes	33.6	30.4	27.8	33.0
None + Nematodes	0	0	0	0
None - Nematodes	16.0	11.9	14.3	16.2

TABLE 5 NO. OF DEVELOPING FRUITS

TREATMENTS	REPLICATES			
	1	2	3	4
2 Fertilizers + Nematodes	0	0	0	0
2 Fertilizers - Nematodes	4	4	5	3
Fertilizer + Nematodes	0	0	0	0
Fertilizer - Nematodes	2	3	2	4
None + Nematodes	0	0	0	0
None + Nematodes	0	0	0	0

APPENDIX G_2

INFLUENCE OF PLANT AGE

TABLE 1 SHOOT WEIGHT (ONE WEEK OLD SEEDLINGS)

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Fertilizer + Nematodes	24.2	19.3	26.4	16.9	21.70
Fertilizer - Nematodes	39.2	43.7	29.6	38.7	37.80
None + Nematodes	9.5	5.7	7.1	8.9	7.80
None - Nematodes	29.2	21.6	28.9	23.7	25.85

APPENDIX G_2

TABLE 2 FRESH ROOT WEIGHT (ONE WEEK OLD SEEDLINGS)

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Fertilizer + Nematodes	12.6	8.4	10.3	9.1	10.10
Fertilizer - Nematodes	10.0	8.6	8.4	10.4	9.35
None + Nematodes	3.3	4.2	3.4	3.9	3.70
None - Nematodes	4.6	6.6	4.9	5.1	5.20

APPENDIX G_2

TABLE 3 FRESH WEIGHT OF TOPS (FOUR WEEK OLD SEEDLINGS)

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Fertilizer + Nematodes	89.6	96.2	81.9	90.7	89.60
Fertilizer - Nematodes	75.6	98.6	79.6	84.9	84.67
None + Nematodes	31.8	24.8	39.6	34.2	32.60
None - Nematodes	49.8	54.2	43.2	59.6	51.70

TABLE 4 FRESH WEIGHT OF ROOTS (FOUR WEEK OLD SEEDLINGS)

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Fertilizer + Nematodes	48.9	40.2	46.4	38.9	43.60
Fertilizer - Nematodes	33.3	38.4	30.6	34.9	34.30
None + Nematodes	14.9	13.2	16.7	16.7	15.38
None - Nematodes	16.9	11.9	14.2	12.6	13.90

EFFECT OF VARIOUS NPK COMBINATIONS

APPENDIX G_3

TABLE 1 FRESH TOP WEIGHTS

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
N- P_1 K_1 + Nematodes	12.7	9.8	15.4	10.9	12.20
N- P_1 K_1 - Nematodes	33.4	39.3	31.9	28.2	33.20
N_1 P- K_1 + Nematodes	18.4	20.6	17.9	13.9	17.70
N_1 P- K_1 - Nematodes	37.8	34.7	39.3	32.6	36.10
N_1 P_1 K- + Nematodes	31.9	21.9	34.9	24.5	28.30
N_1 P_1 K- - Nematodes	39.2	47.8	40.2	49.6	44.20
N_1 P_1 K_1 + Nematodes	38.4	48.9	44.8	33.3	40.10
N_1 P_1 K_1 - Nematodes	47.9	60.5	58.3	43.7	52.60
N_2 P_1 K_1 + Nematodes	44.5	39.6	36.1	47.2	41.85
N_2 P_1 K_1 - Nematodes	53.0	49.2	46.1	58.9	51.80
N_1 P_2 K_1 + Nematodes	37.2	45.3	32.9	42.6	39.50
N_1 P_2 K_1 - Nematodes	55.9	46.2	60.3	51.2	53.40
N_1 P_1 K_2 + Nematodes	36.9	43.7	39.7	38.9	39.80
N_1 P_1 K_2 - Nematodes	48.7	46.2	57.3	48.6	50.20

TABLE 2 FRESH ROOT WEIGHTS

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
N- P_1 K_1 + Nematodes	4.8	6.9	5.7	4.2	6.20
N- P_1 K_1 - Nematodes	7.2	5.1	5.5	7.0	5.40
N_1 P- K_1 + Nematodes	6.4	6.8	8.2	7.2	8.90
N_1 P- K_1 - Nematodes	9.8	8.4	7.3	10.1	7.15
N_1 P_1 K- + Nematodes	9.1	7.7	8.8	8.0	8.40
N_1 P_1 K- - Nematodes	7.9	6.9	7.5	6.5	7.20
N_1 P_1 K_1 + Nematodes	12.5	13.6	10.2	12.9	12.30
N_1 P_1 K_1 - Nematodes	8.9	12.6	11.2	8.1	10.20
N_2 P_1 K_1 + Nematodes	11.6	9.3	11.0	8.9	11.85
N_2 P_1 K_1 - Nematodes	8.9	10.8	10.4	8.3	10.20
N_1 P_2 K_1 + Nematodes	14.2	11.6	16.5	10.9	13.30
N_1 P_2 K_1 + Nematodes	10.6	11.9	13.2	9.1	11.20
N_1 P_1 K_2 + Nematodes	11.2	15.9	13.6	10.9	12.90
N_1 P_1 K_2 - Nematodes	13.4	9.7	12.1	8.4	10.90

APPENDIX G_3

TABLE 3 RATINGS FOR ROOT GALLING AND PERCENTAGE OF ROOTS GALLED

TREATMENTS	RATINGS FOR ROOT GALLING: REPLICATES				% OF ROOTS GALLED: REPLICATES			
	1	2	3	4	1	2	3	4
N $-P_1$ K_1 + Nematodes	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
N_1 P $-K_1$ + Nematodes	5.0	5.0	5.0	5.0	4.0	5.0	5.0	4.0
N_1 P_1 K- + Nematodes	5.0	5.0	5.0	5.0	4.0	5.0	5.0	5.0
N_1 P_1 K_1 + Nematodes	5.0	5.0	5.0	5.0	3.0	3.0	3.0	3.0
N_2 P_1 K_1 + Nematodes	5.0	5.0	5.0	5.0	3.0	3.0	3.0	3.0
N_1 P_2 K_1 + Nematodes	5.0	5.0	5.0	5.0	3.0	3.0	3.0	3.0
N_1 P_1 K_2 + Nematodes	5.0	5.0	5.0	5.0	3.0	3.0	3.0	3.0

APPENDIX H

TABLE 1. EFFICACY OF CARBOFURAN AS INFLUENCED BY TEMPERATURE AFTER INOCULATION

NO. OF NEMATODES PER ROOT SYSTEM TWO WEEKS AFTER INOCULATION

TREATMENTS	TEMPERATURE	REPLICATES		
		1	2	3
+ Carbofuran	27 - 30°C	183	193	166
- Carbofuran	27 - 30°C	410	390	386
+ Carbofuran	18 - 20°C	63	66	73
- Carbofuran	18 - 20°C	376	390	356

TABLE 2 FRESH WEIGHT OF TOPS (6 WEEKS AFTER INOCULATION)

TREATMENTS	TEMPERATURE	REPLICATES				MEAN
		1	2	3	4	
- Carbofuran	27 - 30°C	40.6	48.9	35.2	42.9	41.90
+ Carbofuran	27 - 30°C	62.9	53.2	57.8	50.9	56.20
- Carbofuran	18 - 20°C	46.9	53.9	58.2	49.6	52.15
+ Carbofuran	18 - 20°C	34.2	27.8	21.8	32.7	29.10

APPENDIX H

TABLE 3 FRESH WEIGHT OF ROOTS

TREATMENTS	TEMPERATURE	REPLICATES				MEAN
		1	2	3	4	
− Carbofuran	27 - 30°C	14.2	13.1	17.6	15.9	15.20
+ Carbofuran	27 - 30°C	12.6	14.2	11.9	12.5	12.05
− Carbofuran	18 - 20°C	14.6	12.2	14.8	11.1	13.18
+ Carbofuran	18 - 20°C	11.9	9.8	10.1	10.6	10.60

TABLE 4 NO OF GALLS PER ROOT SYSTEM

TREATMENTS	TEMPERATURE	REPLICATES				MEAN
		1	2	3	4	
- Carbofuran	27 - 30°C	1014	899	1130	1045	10220
+ Carbofuran	27 - 30°C	524	411	484	589	5020
- Carbofuran	18 - 20°C	924	732	791	769	8040
+ Carbofuran	18 - 20°C	282	200	221	239	2355

APPENDIX I

TABLE 1 ORGANIC MATTER CONTENT OF SOIL

NEMATODES PER ROOT SYSTEM 14 DAYS AFTER INOCULATION

TREATMENTS	REPLICATES			MEAN
	1	2	3	
Sand + Carbofuran	130	123	106	119.7
Sand - Carbofuran	430	400	456	428.7
20% Organic Matter + Carbofuran	116	140	130	128.7
25% Organic Matter - Carbofuran	406	430	413	416.3
50% Organic Matter + Carbofuran	256	270	280	268.7
50% Organic Matter - Carbofuran	283	320	326	309.7

APPENDIX I

TABLE 2 FRESH WEIGHT OF SHOOT

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Sand + Carbofuran	68.9	57.2	62.7	66.8	63.90
Sand - Carbofuran	52.9	44.2	50.1	39.4	41.60
25% Organic Matter + Carbofuran	58.6	66.9	51.6	66.1	66.80
25% Organic Matter + Carbofuran	40.3	46.1	37.8	42.2	46.65
50% Organic Matter + Carbofuran	55.9	58.2	62.4	50.7	56.80
50% Organic Matter + Carbofuran	47.8	56.7	51.1	42.8	49.60

APPENDIX I

TABLE 3 FRESH WEIGHT OF ROOTS

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Sand + Carbofuran	12.6	13.3	12.4	14.9	10.30
Sand - Carbofuran	11.6	9.9	10.6	9.1	13.30
25% Organic Matter + Carbofuran	12.0	13.2	9.2	10.4	11.20
25% Organic Matter - Carbofuran	13.8	16.1	13.6	12.9	14.20
50% Organic Matter + Carbofuran	13.6	12.2	14.0	11.8	12.90
50% Organic Matter - Carbofuran	15.2	15.2	16.3	14.0	15.18

APPENDIX I

TABLE 4 NO. OF GALLS PER ROOT SYSTEM

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Sand + Carbofuran	356	307	402	319	346.0
Sand - Carbofuran	1114	1054	992	1326	1121.5
25% Organic Matter + Carbofuran	404	486	416	394	425.0
25% Organic Matter - Carbofuran	913	1089	896	1046	986.0
50% Organic Matter + Carbofuran	690	804	700	702	724.0
50% Organic Matter - Carbofuran	787	796	801	787	792.8

APPENDIX J

EFFECT OF SINGLE AND MULTIPLE APPLICATION

TABLE 1 FRESH SHOOT WEIGHT

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Control Fertilizer only	68.4	58.4	66.9	54.1	61.95
8 kg ai/ha at transplanting	89.2	76.6	80.4	84.2	82.60
4 + 4 (2 weeks) after transplanting	79.6	83.2	73.1	72.9	77.20
8 + 4 (2 weeks) after "	80.4	70.6	81.2	68.4	75.15
8 + 4 (3 weeks) after "	97.9	78.1	84.2	92.6	83.70

APPENDIX J

TABLE 2 ROOT WEIGHT

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Fertilizer only	44.8	37.3	48.3	40.2	42.65
8 kg ai/ha at transplant	30.5	27.8	25.1	30.1	28.38
4 + 4 (Split) 2 weeks	28.2	30.7	25.9	22.9	26.93
8 + 4 2 weeks	18.5	23.9	18.2	21.8	20.60
8 + 4 8 weeks	28.1	22.8	23.7	26.2	25.20

TABLE 3 NO. OF GALLS PER ROOT SYSTEM

TREATMENTS	REPLICATES				MEAN
	1	2	3	4	
Fertilizer only	2958	3420	2862	2872	3028
8 kg ai/ha at transplant	1020	937	816	1075	962
4 + 4 (split) 2 weeks	1120	989	1090	941	1035
8 + 4 2 weeks	425	399	356	484	416
8 + 4 3 weeks	563	496	551	506	529