

ROOT-KNOT NEMATODE CONTROL INVESTIGATIONS IN TOBACCO IN QUEENSLAND

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SUMMARY.

During 1948-55, 8 field trials were set out in North Queensland to investigate the control of the root-knot nematode *Meloidogyne javanica* (Treub) Chitwood, which is a serious pest of tobacco.

It is shown that, irrespective of the use of nematocides in the field, even a trace of nematode galling in seedlings can be primarily responsible for appreciable economic losses.

In heavily infested irrigated fields, yields and appraised values can be increased more than threefold by the correct use of either DD or EDB.

Where tobacco is grown each year on the same site, galling tends to increase at a greater rate than where resistant crops are used in the rotation. This use of resistant crops, however, does not prevent consistently an increase in galling; one of the reasons may be the impracticability of obtaining pure stands of resistant plants.

I. INTRODUCTION.

The recognised problems with root-knot nematode as a pest of tobacco in Queensland have been the complete protection of seedlings and the protection of field plants at least during establishment and the early growth stages. Most of the earlier literature on tobacco pests, including nematodes, is consolidated in articles by J. H. Smith (1931, 1933), Veitch (1938, 1942), Officers of the Department of Agriculture and Stock (1951) and W. A. Smith (1952).

Lamb (1892) and subsequent authors recommended firing for nematode control in seedbeds and the method has been the commercial practice for over half a century. Haney (1950), following American thought at that time, tested chemical methods for use in seedbeds. The results indicated that when the three essentials of weed control, plant growth and nematode control were considered, firing was still the best treatment. In view of the continued general success of firing, no further major effort was made to screen chemicals in seedbeds. For future guidance, however, a trial was carried through to the field to demonstrate the effect of seedling infestation on yields.

As effective nematocides were not available until recent years, recommendations for nematode control in the field were necessarily restricted to maintaining plant vigour by cultivation and fertilizing, uprooting crop residues after harvesting, rotating with immune crops, building up the soil organic matter, and eliminating chance pest distribution. All these practices may have helped in some degree with crop production in infested land, but none has been proved reliable.

The field investigations in North Queensland recorded in this paper were carried out during the period 1948-1955, principally at Claredale in the Lower Burdekin district. These consisted of seven trials with nematocides, the examination of root infestation in a crop rotation trial (see Appendix), and observations on commercial control.

During the 1950-51 season in the Bundaberg and Miriam Vale districts, C. D. Brickhill set out field trials to test DD levels and placement positions, and found that commercial nematode control was possible in rain-grown tobacco when treatment was restricted to the row position (unpublished reports, Queensland Department of Agriculture and Stock).

II. SPECIES INVOLVED.

Prior to 1954, root-knot nematode was referred to by Queensland authors first as *Heterodera radicola*, then as *Heterodera marioni* (Cornu) Goodey. Smith (1954), anticipating Colbran (1955), used the name *Meloidogyne* spp. Recent root-knot nematode surveys by Colbran (unpublished reports, Queensland Department of Agriculture and Stock) show that only *Meloidogyne javanica* (Treub) Chitwood has been recorded from tobacco in northern Queensland, and this is the species dealt with in this paper.

III. MATERIALS.

The nematocides used were:—

DD.—A commercial mixture of 1,3 dichloropropene and 1,2 dichloropropene.

EDB.—Ethylene dibromide: used in power kerosene, 1 pint EDB made up to 1 gallon (EDB (Sp. G. $\frac{2.5}{2.5}$ 2.170) content 12.5 per cent. v/v).

CBP.—Chlorobromopropene.

IV. METHODS.

(1) General.

For the trial with infested seedlings a seedbed 20 ft. x 4 ft. was fired correctly at one end, grading off to an ineffective burn at the other. Seedlings were hosed out, and the roots floated in water for grading into the categories required. Only clean seedlings from fired beds were used in the other trials.

All the field trial plantings were watered by furrow irrigation. Precautions were taken during land preparation to prevent excessive movement into treated parts of soil which might contain nematodes and also within plots where nematocide placement was restricted. In the 1948-49 screening trial at Mareeba, DD with and without an emulsifier was applied in the irrigation

water and by injection. In subsequent trials only the injection method was used. A light commercial hand injector metering fumigant by piston strokes from a $\frac{1}{2}$ -gal. tank provided reasonable accuracy in treating the field plots. Strings marked at intervals of 1 ft. were used as a guide during injection. The depth of injection was determined by a stop-plate fixed 6 in. above the spear outlet holes.

All experimental tobacco was grown in rows 4 ft. apart, and treatments were made 2-3 weeks before planting out. Three types of placement were used—the "4 line", requiring injection lines 6 in. and 18 in. each side of the row position; the "2 line", 6 in. each side; and the "1 line", on the row position.

The dosage rates used in successive trials varied until 20 gal. per acre was selected as a standard. The rates per acre actually used with the equivalent rates per foot are shown in Table 1.

Table 1.
NEMATOCIDE DOSAGE RATE EQUIVALENTS.

gal./ac.	ml./ft.		
	4 line.	2 line.	1 line.
33.3	3.5
20.0	2.1
19.1	2.0
16.7	1.8
10.0	..	2.1	4.2
9.6	1.0
8.1	..	1.7	..

No water-sealing of injected plots was practised except in the 1948-49 screening trial, when a watering can was used for the purpose.

Soil temperatures during treatment in all trials were mild and no adverse effects due to this factor were expected or noticed. Control of insect pests, according to current Departmental recommendations, was implemented as required in all trials.

Layout, plot size, details of the various treatments and other relevant information are given with the results for each trial.

(2) Assessing Results.

(a) Yields.

Yields from the 1950-51 Claredale trials were taken as weight of harvested green leaf per plant. Stand counts were made and the leaf was picked as it ripened.

It was found that relative green weights may vary more than the corresponding cured leaf weights, so yields from the later trials are expressed in economic units. As the crop ripened, plots were harvested separately and labelled sticks of leaf were cured and afterwards bulked in parcels. When ready for grading, the parcels were weighed and recorded by plots. The sum of grade weights is given in the 1953-54 trial results as the yield of cured leaf after grading. This differs from the yield before grading by variation in moisture content and loss of trash.

Samples of each grade were given an assessed value by a trade appraiser, and relevant plot values were calculated accordingly. The appraised value is the only criterion which takes into account the effects of nematode infestation on quality as well as on quantity of tobacco leaf.

(b) Root-Knot Indices.

The method used to assess the degree of root galling, usually after harvesting, is essentially that of Smith and Taylor (1947). The root-knot index for a plot, calculated after rating the roots of each plant, is an estimate of percentage infestation.

V. RESULTS.

(1) Field Trial with Infested Seedlings.

The categories used to distinguish nematode infestations of seedlings were:—

Clean—free from galls.

Trace—with one or two minute galls.

Distinct—with one or two distinct galls.

Moderate—with distinct galling distributed over several roots.

Table 2.

YIELDS AND APPRAISED VALUES.
(Field trial with infested seedlings.)

Soil Treatment.	Seedling Infestation.	Yield of Cured Leaf Before Grading (lb./ac.).	Appraised Value (£/ac.).
	Clean	992	449
	Trace	702	274
	Distinct	632	304
	*Moderate
DD 20 gal./ac. 4 line	Clean	1,700	976
DD 20 gal./ac. 4 line	Trace	1,652	870
DD 20 gal./ac. 4 line	Distinct	1,260	631
DD 20 gal./ac. 4 line	Moderate	578	286
Necessary differences	} 5% } 1%	277	107
for significance		374	144

* This treatment was not included in the analyses because of shortage of seedlings of the correct rating.

This trial was planted out on Oct. 5, 1954, as a (4 x 2) x 5 randomised block with a plot size of 2 rows x 47 ft., adjacent to the 1954-55 nematocide trial at Claredale. Yields and appraised values are given in Table 2. Root-knot indices were not obtained, as fungal breakdown following heavy rains soon after harvesting caused severe root destruction.

Differences in plant growth were not apparent until the approach of harvesting. At that time the degree of wilting present was inversely related to the yields obtained.

(2) 1948-49 Screening Trial, Mareeba.

The light-grey sandy soil had grown tobacco two years previously. Plot size was 4 rows x 32 ft. in a 5 x 5 randomised block layout. Planting was carried out on Oct. 23-25. Root-knot indices only, taken from 20 plants in the two inner rows of each plot, were obtained; they are given in Table 3.

Table 3.

ROOT-KNOT INDICES.
(1948-49 Screening Trial, Mareeba.)

Treatment.	Mean Root-Knot Index.					
DD injected 33.3 gal./ac. 4 line	7.2					
DD injected 16.7 gal./ac. 4 line	25.6					
DD with emulsifier in irrigation water 16.7 gal./ac. ..	38.8					
DD in irrigation water 16.7 gal./ac.	47.0					
Control	30.4					
Necessary difference for significance	<table style="display: inline-table; vertical-align: middle;"> <tr> <td rowspan="2" style="font-size: 2em; vertical-align: middle;">}</td> <td>5%</td> <td>18.5</td> </tr> <tr> <td>1%</td> <td>25.5</td> </tr> </table>	}	5%	18.5	1%	25.5
}	5%		18.5			
	1%	25.5				

The DD when run into the irrigation water settled on the bottom of the furrows and probably evaporated when the water soaked in.

(3) 1950-51, Claredale.

Three sites were used on the light-textured soils of the Burdekin association. Plot size was 2 rows x 36 ft., and each trial was a 4 x 6 randomised block. Two of these were to test row treatments simultaneously with Brickhill's work in southern Queensland, and the third to check the application of DD at the lower strength which had failed at Mareeba the previous season.

The trials, planted during late September and early October, were subjected to unseasonable heavy rains which caused some waterlogging and reduced plot yields. The results are given in Tables 4 and 5.

Table 4.

YIELDS.
(1950-51, Claredale.)

Treatment.	Green Leaf Yields (lb./plant).		
	Trial I.	Trial II.	Trial III.
DD 19.1 gal./ac. 4 line	1.46	1.14	1.26
DD 8.1 gal./ac. 2 line	1.31	1.12	..
DD 9.6 gal./ac. 4 line	1.21
Control	1.27	0.88	0.94

Differences are not significant.

Table 5.

ROOT-KNOT INDICES.
(1950-51, Claredale.)

Treatment.	Mean Root-Knot Indices.		
	Trial I.	Trial II.	Trial III.
DD 19.1 gal./ac. 4 line	26.0	22.2	9.4
DD 8.1 gal./ac. 2 line	66.9	59.4	..
DD 9.6 gal./ac. 4 line	29.5
Control	84.2	86.8	85.1
Necessary differences for significance	5%	25.2	17.8
	1%	35.8	25.2
			11.6
			16.4

The efficacy of DD in preventing root galling was not reflected in the yields, probably due to the weather factor. Overall yields were low.

(4) 1953-54, Claredale.

A 6 x 5 randomised block layout was used, with a plot size of 2 rows x 70 ft. The soil type was similar to that for Trial I, 1950-51, the two blocks being adjacent.

Planting out took place during late September, and early difficulties in establishment led to some unevenness in plant stand. The results are given in Table 6.

Untreated plants were not as tall as the others but the difference was not outstanding and was not apparent before flowering. Mild wilting and premature ripening of leaf were noticeable in the control plots from about the time of the first main flowering.

Table 6.

YIELDS, APPRAISED VALUES AND ROOT-KNOT INDICES.
(1953-54, Claredale.)

Treatment.	Mean Yield Cured Leaf After Grading (lb./ac.).	Appraised Values (£/ac.).	Mean Root-Knot Index.
EDB 20 gal./ac. 4 line	1,482	722	37.5
EDB 10 gal./ac. 2 line	1,569	747	46.1
DD 10 gal./ac. 2 line	1,460	662	58.3
DD 20 gal./ac. 4 line	1,619	789	63.0
DD 10 gal./ac. 1 line	1,538	707	71.5
Control	826	313	96.8
Necessary differences for significance	5%	119	16.7
	1%	162	22.8

The tolerance of the tobacco plant under irrigated conditions to moderate root-knotting as indicated by the indices is reflected in these results. The value of the nematocides, and of the 2 line and 1 line placements, is obvious when nematode infestation is severe.

(5) 1954-55, Claredale.

This trial was planted in September on Burdekin fine sand which had carried a heavily infested crop during the first half of the year. A 6 x 4 randomised block layout with a plot size of 2 rows x 84 ft. was used, with control strips of 2 rows x 80 ft. through the block in place of randomised control plots. Results are given in Table 7.

Table 7.

YIELDS, APPRAISED VALUES AND ROOT-KNOT INDICES.
(1954-55, Claredale.)

Treatment.	Mean Yield Cured Leaf Before Grading (lb./ac.).	Appraised Value (£/ac.).	Mean Root-Knot Index.
EDB 20 gal./ac. 4 line	1,979	811	19.4
EDB 10 gal./ac. 2 line	1,639	718	22.9
EDB 10 gal./ac. 1 line	1,610	673	24.7
DD 20 gal./ac. 4 line	1,552	666	25.6
DD 10 gal./ac. 2 line	1,499	651	29.6
DD 10 gal./ac. 1 line	1,105	469	59.4
Control	(675)	(196)	(74.0)
Necessary differences for significance	5%	207	16.2
	1%	286	22.2

No early stunting was observed in the untreated plants. During harvesting the leaves ripened rapidly and prematurely, and frequently wilted before picking.

(6) Screening of CBP.

During the 1954-55 season at Claredale a 4 x 5 randomised layout with control strips was used to screen CBP against DD as the standard. Plot size was a single row of 10 plants.

Plants were pulled for root-knot assessment six weeks after planting out. The indices given in Table 8 are therefore not comparable with those figures from the other trials.

Table 8.

ROOT-KNOT INDICES.
(Screening of CBP.)

Treatment.	Mean Root-Knot Index.
DD 20 gal./ac. 4 line	0.0
CBP 10 gal./ac. 4 line	2.6
CBP 15 gal./ac. 4 line	1.1
CBP 20 gal./ac. 4 line	6.9
Control	(13.3)
Necessary difference for significance 5%	3.2

The nature of the variation indicates an inadequacy in the small-plot technique. CBP, however, does not give promise of improving on DD, and is a pungent liquid most unpleasant to handle.

VI. DISCUSSION.

Results from the field trial with infested seedlings clearly demonstrate (Table 1) that only clean seedlings should be planted out. Irrespective of the use of nematocides in the field, even a trace of nematode galling in tobacco seedlings can be primarily responsible for appreciable economic losses.

A high percentage of Queensland tobacco land is infested in some degree by root-knot nematodes, and there has been a tendency to throw the worst of this land out of tobacco production. No ready method of sampling the pest populations at the time of planting, to help predict nematode damage in the subsequent crop, was available to the author. In commercial practice, district and field histories are used for this purpose. All trial sites had recent histories of heavy infestations and the results show that under these circumstances the use of nematocides in irrigated crops can increase yields, as appraised values, more than threefold (Table 7). Concurrent with this work, soil fumigating

machines were evolved, mostly by local effort, for translating experimental results into the commercial sphere. W. A. Smith (1954) first firmly recommended the field use of nematocides, and again the next year as follows:—

“... fumigate the soil when free from weeds and in good tilth with EDB or DD at least three weeks before planting out. The recommended dosages per acre are 10 gal. of 12½% EDB or 10 gal. of DD. Fumigation depth should be 6 inches, with thorough sealing after application.

Where careful soil working can be achieved during the early life of the crop, the intended planting line may be treated by a single application along the centre of the row, or one 6 inches each side of it. The double line treatment makes allowance for the lateral movement of soil in and near the planting lines during working. Treatments in rows 1 foot apart over the whole field, requiring 20 gal. of fumigant per acre, permit maximum lateral working of soil, but if reasonable care is taken this more expensive treatment is unnecessary.”

“Soil fumigants and detailed dosage rates are:—

(1) *EDB* (12½% concentration).

Machine Application.

Treatment in the centre of row position—1 pint to $2\frac{1}{16}$ chains (136 feet),

or,

Treatment 6 inches each side of row position—1 pint from each outlet to $4\frac{1}{8}$ chains (272 feet). The same rate applies if 20 gallons are used in rows 1 foot apart across the field.

Hand Injector Application.

Same positions as for machine application but with injections 1 foot apart. Rate: 1 fluid ounce per each 7 injections for centre of row treatment, and 14 injections totalling 1 fluid ounce for the other treatments.

(2) *DD.*

Use the commercial material at the same rates as for 12½% EDB.”

At present most tobacco in Queensland is irrigated, and the conclusion from general field observations is that increased yields, comparable with experimental results, are obtainable when strict attention is given to control details as recommended above.

VII. ACKNOWLEDGEMENTS.

Facilities for conducting the trials were provided by the Agriculture Branch of the Department of Agriculture and Stock, and by several farmers. Mr. G. W. Saunders (Assistant Entomologist) was in charge of the 1954-55 trials and other Departmental officers assisted with all trials. The statistical analyses were carried out by Mr. P. B. McGovern (Biometrician, Division of Plant Industry).

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APPENDIX.

Root-Knot Nematode in a Crop Rotation Trial.

A crop rotation trial on the Clare Tobacco Experiment Farm, Claredale, provided an opportunity to record the extent of root galling on tobacco in six successive years. The land was open forest on the Burdekin levee, cleared and prepared in the first half of 1948. It had previously been used as grazing land. The trial was arranged as an 8 x 4 randomised block with a plot size of 8 rows x 56 ft.

Appendix Table 1.

ROOT-KNOT INDICES.

(Crop Rotation Trial.)

Rotation.	Year.					
	1948.	1949.	1950.	1951.	1952.	1953.
1	5.0	12.6	64.0	50.7	85.0	79.3
2	5.4	9.7	66.7	42.6	75.1	49.6
3	2.8	9.6	32.8	49.0	75.8	88.5
4	13.7	67.6
5	5.5	..	29.1	27.7
6	6.1	22.2	62.5	..
7	14.0	..	26.2	..	49.6	..
8	10.2	38.4	..	43.6	..	66.3

The year was divided into three equal cropping periods. Before each tobacco crop the land was bare-fallowed for one period. Other crops in the annual series of rotations were volunteer weeds (Table 1, Rotation 1), Gambia pea (*Crotalaria goreensis* Guill. & Perr.) (Rotation 2), and peanuts (*Arachis hypogaea* L.) (Rotation 3). A cycle of Gambia pea, potato (*Solanum tuberosum* L.) and 20 months of Rhodes grass (*Chloris gayana* Kunth) preceding tobacco was repeated in three time phases (Rotations 4, 5, 6). Twelve months of Gambia pea and 4 months of maize (*Zea mays* L.) was repeated in two time phases (Rotations 7, 8).

In plant species composition surveys towards the end of April in 1950, 1952 and 1953 it was found that all plots carried a variety of weeds either as a trace or in larger numbers. The Rhodes grass plots carried fewer weeds than most others.

Each tobacco crop was pulled after harvesting, and root-knot indices were assessed. The results are given in Appendix Table 1.

Despite wide variation in behaviour between plots, a general increase in root-knot galling with tobacco cropping was demonstrated. None of these variations could be readily explained from general observations, but broad conclusions may be drawn from the data in Table 1. Where tobacco is grown each year on the same site, galling tends to increase at a greater rate than where resistant crops are used in a rotation. This use of resistant crops, however, does not prevent consistently an increase in galling: one of the reasons may be the impracticability of obtaining pure stands of resistant plants.
