

NN 8201

no 491

C

THE EFFECT OF PARTIAL SOIL  
STERILIZATION ON PLANT PARASITIC  
NEMATODES AND PLANT GROWTH

M. F. M. EISSA

NN08201.491

BIBLIOTHEEK  
DER  
NDBOUWHOGESCHOOL  
EN. FOULKESWEG 1a  
WAGENINGEN

**THE EFFECT OF PARTIAL SOIL STERILIZATION ON PLANT  
PARASITIC NEMATODES AND PLANT GROWTH**

Dit proefschrift met stellingen van

MOHAMED FAHMI MOHAMED EISSA,

M. Sc. (Nematology), University of Cairo, Egypte, geboren te Zagazig, Egypte, 2 januari 1942, is bewerkt onder leiding van Dr. Ir. M. Oostenbrink, lector in de Nematologie, en is goedgekeurd door de promotor, Dr. Ir. J. DEKKER, hoogleraar in de Fytopathologie.

*De Rector Magnificus van de Landbouwhogeschool,*

J. M. POLAK

*Wageningen, 8 april 1971*

**THE EFFECT OF PARTIAL SOIL  
STERILIZATION ON PLANT PARASITIC  
NEMATODES AND PLANT GROWTH**

*(with a summary in Dutch)*

**PROEFSCHRIFT**

**TER VERKRIJGING VAN DE GRAAD  
VAN DOCTOR IN DE LANDBOUWWETENSCHAPPEN  
OP GEZAG VAN DE RECTOR MAGNIFICUS, MR. J. M. POLAK,  
HOOGLERAAR IN DE RECHTS- EN STAATSWETENSCHAPPEN  
VAN DE WESTERSE GEBIEDEN  
TE VERDEDIGEN TEGEN DE BEDENKINGEN VAN EEN  
COMMISSIE UIT DE SENAAAT  
VAN DE LANDBOUWHOGESCHOOL TE WAGENINGEN  
OP WOENSDAG 2 JUNI 1971 TE 16.00 UUR**

**DOOR**

**MOHAMED FAHMI MOHAMED EISSA**

**This thesis is also published as Mededelingen Landbouwhogeschool Wageningen 71-14 (1971)  
(Communications Agricultural University Wageningen, The Netherlands)**

## THEOREMS

### I

Old cultivated soils are usually unhygienic substrates for plants and partial soil sterilization should become a common agricultural technique.

(This Thesis)

### II

Instead of a soil fungicide, a biocide with a wider spectrum of biological activity should be preferred for control of a soil-borne fungal disease.

(This Thesis)

### III

The specificity of tobacco rattle virus transmission by *Trichodorus pachydermus* is probably determined by the charge of the surface of the virus particles and that of the cuticle of the digestive tract of the nematode.

(C. E. TAYLOR & W. M. ROBERTSON, J. Gen. Virol. 6 (1970): 179-182)

### IV

Chemical control will remain the major method for pest control as long as alternative methods are still neither practical nor economical.

### V

In U.A.R. (Egypt), the high-dam project will increase the national agricultural income despite lowering the soil fertility.

(H. ABDALLAH, U.A.R. Agriculture. Foreign Relations Dept., Minis. Agric. 1965: 115 p.)

(ANONYMOUS, Statistical Abstract of the United Arab Republic. Central Agency for Public Mobilisation and Statistics, Cairo 1970: 182 p.)

### VI

In developing countries, a wider basis in educational pyramids is necessary for better progress in the industrial as well as in the agricultural fields.

## **VII**

**In arid lands, breeding for drought and salinity resistance especially for orchard plantations, could be of more value than breeding for disease resistance.**

## **VIII**

**Industrialization of raw materials in the developing exporting countries could be a means for obtaining better human nutrition in these countries as a result of economic improvement.**

**'Micro-organisms are the eventual heirs to the dead bodies of all animals, including those of micro-biologists'.**

**S. D. GARRETT, 1964**

*To every constructive hand in human welfare,  
To all those who contributed in my education and those who would,  
To my parents, wife and son.*



# CONTENTS

<b>1. INTRODUCTION</b>	<b>1</b>
1.1. Partial soil sterilization (PSS) and estimation of crop losses	1
1.2. Scope of the study	2
<b>2. LITERATURE REVIEW</b>	<b>3</b>
2.1. Physical means	3
2.1.1. Heat	3
2.1.2. Freezing	3
2.1.3. Drought	3
2.1.4. Flooding	4
2.1.5. Electricity	4
2.1.6. Irradiation	4
2.1.7. Mechanical disturbance	4
2.2. Chemical means	4
2.2.1. Early nematicides	4
2.2.2. Broad-spectrum soil fumigants	5
2.2.3. Halogenated hydrocarbons	7
2.2.4. Methylisothiocyanate compounds (Trapex and Vapam)	10
2.2.5. Organophosphorous compounds and other systemics	11
2.2.6. Soil fungicides	11
2.2.7. Other soil pesticides	12
2.3. PSS effects	12
2.3.1. Elimination of noxious organisms	13
2.3.2. Effect on soil fertility	14
2.3.3. Direct influence of soil pesticides on plants	15
<b>3. MATERIALS AND METHODS</b>	<b>17</b>
3.1. Inventory of data on PSS effects	17
3.2. Field experiments, glasshouse and laboratory studies	17
3.3. Nematicides and their application	17
3.4. Test plants and their evaluation	18
3.5. Nematodes and their enumeration	18
3.6. Statistical treatment of the data	18
<b>4. INVENTORY OF QUANTITATIVE DATA</b>	<b>19</b>
4.1. Introduction	19
4.2. Inventory	19
4.3. Discussion	24
4.3.1. General averages	24
4.3.2. Treatment averages	29
4.3.3. Crop averages	36
4.3.4. PSS results related to the presence of parasitic organisms	43
4.3.5. PSS results in relation to soil type	48
4.3.6. PSS results in relation to locations	49
<b>5. PSS ON FIELDS WITHOUT KNOWN PROBLEMS</b>	<b>51</b>
5.1. Introduction	51
5.2. Experiments	51
5.3. Discussion	54

6. PSS OF SOILS WITH KNOWN NEMATODE PESTS . . . . .	56
6.1. Introduction . . . . .	56
6.2. Sandy soil with <i>Heterodera rostochiensis</i> under potato . . . . .	57
6.3. Clay soil with <i>Heterodera goettingiana</i> under pea . . . . .	61
6.4. Sandy polder soil with <i>Hemicycliophora conida</i> under grass . . . . .	64
6.5. Sandy glasshouse soil with <i>Meloidogyne javanica</i> under tomato . . . . .	67
6.6. Polder clay soil with <i>Meloidogyne naasi</i> under cabbage . . . . .	69
6.7. Polder clay soil with <i>Meloidogyne naasi</i> under barley . . . . .	71
6.8. Sandy polder soil with <i>Meloidogyne naasi</i> under barley . . . . .	73
6.9. Loamy sand (PD) soil with a complex of <i>Pratylenchus crenatus</i> , <i>Tylenchorhynchus dubius</i> and <i>Rotylenchus robustus</i> under oat . . . . .	74
6.10. Discussion . . . . .	76
7. PSS TREATMENTS OF FRESH OR UNINFESTED SOILS OF DIFFERENT TYPES . . . . .	79
7.1. Introduction . . . . .	79
7.2. Effect on carrot . . . . .	81
7.3. Effect on rose . . . . .	86
7.4. Effect on ryegrass . . . . .	91
7.5. Discussion . . . . .	94
8. DIAGNOSTIC STUDIES ON AN UNIDENTIFIED PSS EFFECT IN OAT . . . . .	95
8.1. Introduction . . . . .	95
8.2. Experiments . . . . .	95
8.2.1. Introductory experiment 1968 . . . . .	95
8.2.2. Experiments in 1969 . . . . .	99
8.2.3. Effect of the inoculation procedure on <i>P. crenatus</i> , <i>T. dubius</i> and <i>R. robustus</i> . . . . .	108
8.3. Discussion . . . . .	110
9. SUMMARY . . . . .	112
10. SAMENVATTING . . . . .	115
11. ACKNOWLEDGEMENTS . . . . .	118
12. REFERENCES . . . . .	119

# I. INTRODUCTION

## 1.1. PARTIAL SOIL STERILIZATION (PSS) AND ESTIMATION OF CROP LOSSES

Partial soil sterilization is any physical or chemical soil treatment which strongly reduces certain groups of soil organisms.

The physical methods comprise treatments by heat, freezing, drought, flooding, electricity, radiation, or mechanical disturbance of the soil. The chemical methods comprise various treatments of the soil, or sometimes of the crops, with pesticides. PSS is a powerful technique to eliminate yield deficiencies in crop production and may help to improve the food supply in the world as a whole (VALLEGA and CHIARAPPA 1964). It is applied in agriculture to compensate yield losses due to soil-borne pests and pathogens and unknown causes in cultures of high value and would no doubt find wide application in many field crops if costs were not prohibitive. Numerous results of specific treatment – crop – field combinations are published, and reports are available on wide-scale application of particular treatments in certain areas. The literature on soil disinfection, especially on the application of nematicides, is already vast and is increasing rapidly.

In the years 1965 up to 1970 about 400 articles on soil disinfection have been published, which is 13% of the total nematological literature, according to the card file by BAKER, OOSTENBRINK and VAN BERKUM (1967 onward).

In The Netherlands at least 30,000 ha of agricultural land was treated with 250 l of dichloropropene formulations in 1969 (HIJINK, pers. comm.). Also the flower-bulb soils (about 12,000 ha) and the glasshouse soils (about 5,000 ha) are usually treated once every 2 or 3 years, as well as part of the vegetable cultures in the field and of replanted orchards, nurseries for woody plants, ornamentals, strawberries and some other crops. In these cases higher doses and more expensive materials are often used than for agricultural land. It is a conservative estimate that in The Netherlands in 1969 more than 40,000 ha, about 4% of the arable land, were treated with soil disinfectants and that the value of the materials exceeded 10 million guilders. In the U.S.A. over half a million acres for growing high value crops, such as tobacco, vegetables and pineapples have been treated in 1959 (TAYLOR 1961).

GOOD and FELDMESSER (1966) reported that one million acres were treated annually with soil disinfectants representing a sales value of 25 million dollars. This area has increased since then: in 1969 about one and a half million acres, which still is less than 1% of the cultivated land, was treated (GOOD, pers. comm.).

Despite many publications on soil disinfectants, reliable estimates about the potential overall effect of PSS on agricultural production are scarce, and this holds for specific treatments on specific crops.

TAYLOR (1961) reviewed 853 experiments and reported a yield increase for treated soil as compared to untreated of 36% for lima beans, 51% for corn, 91% for cotton, 90% for okra, 126% for soybeans, 175% for sugarbeet, 13% for tobacco and 73% for tomatoes. The general increase for all crops was 87%. Several authors pointed out that the yield increase could not be attributed only to the control of nematodes. TAYLOR stated that the contribution of other soil organisms is difficult to assess, but did not exceed 10% of the value obtained in his experiments. TAYLOR's results, however, were obtained on fields with known problems and are therefore not representative for normal farm soils.

There are, on the other hand, a number of estimates of the damage caused by nematodes, and these losses are compensated by PSS at any rate. An overall loss of more than 10% is considered an appropriate estimate by several authors (LEAP 1955, OOSTENBRINK 1957, TAYLOR 1967 and GOOD, pers. comm.). HUTCHINSON (1959) estimated U.S. \$ 15 million as the yearly loss for New Jersey and ALLEN and MAGGENTI (1959) indicated more than \$ 90 million for California. HOLLIS (1962) suggested the possibility of a 50% increase in crop production if nematode pests would be controlled on Kenya farms. Losses estimated as 10% of the total crops value in the U.S.A. would reach 4-5 thousand million dollars (BROWN 1963). Other estimates for the U.S.A. are lower: \$ 500 million (CAIRNS 1955); \$ 250 million (HUTCHINSON et al. 1961); or \$ 372 million on an average for field, fruit, nut and vegetable crops in U.S.A. for the years 1951-1960 (LE CLERG 1964). OTEIFA (1965) estimated that the yearly loss in Egyptian horticultural crops due to plant nematodes was £ 2 million. SOUTHEY and SAMUEL (1954) reported a loss of £ 2 million for potato in England and Wales due to *Heterodera rostochiensis*.

Knowledge of the total damage caused by nematodes and other organisms in the soil is incomplete. At any rate the present estimate is lower than the total PSS effect. How far this gap is due to underestimating the damage by either soil-borne pests and diseases, or other effects of PSS, which are known to exist, cannot be indicated on the basis of present knowledge.

## 1.2. SCOPE OF THE STUDY

The purpose of this study was to get quantitative information about the power of PSS to increase crop yields in The Netherlands, and the main parameter used to determine this power was crop yield from treated as compared to untreated soil. An extensive inventory of published and unpublished data was made. Experimental work under field and laboratory conditions, in which emphasis was laid on nematode infestation, was added. The study dealt with the situation in The Netherlands, where nowadays more soil disinfectants are used per ha cultivated land or per inhabitant than in other countries, including the U.S.A.

## 2. LITERATURE REVIEW

Two files were the basis for literature study on partial soil sterilization in general and on soil sterilization against plant nematodes in particular. They were file of summaries on 'Chemical control of plant nematodes', and supplements, published by PEACHY and CHAPMAN (1966, 1967 and 1968) and the literature file on 'Plant, soil and freshwater nematodes', published and currently complemented by BAKER, OOSTENBRINK and VAN BERKUM (1967 onwards). The literature has been arranged according to the means of PSS as follows:

### 2.1. PHYSICAL MEANS

#### 2.1.1. Heat

Steaming soil is a widespread method of PSS in horticulture. It was reported early by RUSSEL and BUDDIN (1914). MELCHERS (1919) protected glasshouse vegetables against root-knot infestation by using steamed soil. TRIFFITT and HURST (1935) recorded strong influence of soil humidity on the effect of heat treatment. JOHNSON (1946) warned against secondary, harmful effects caused by steaming the soil. A general treatise on the application of heat for soil sterilization was given by LAWRENCE (1956). A new development is the application of steam mixed with air, which is effective although the temperature reached in the soil is less than 100°C (BAKER and OLSEN 1960). Finally: dry heat, generated by passing electricity through wires in the soil (VAN DEN BRANDE and GILLARD 1957) is also a method of PSS, as well as burning wood to prepare soil for the cultivation of pepper as described by VAN DER VECHT (1950).

#### 2.1.2. Freezing

Plant nematodes can normally endure the low temperatures which occur in their natural habitat and are rather preserved than killed by it. Frost stops nematode activity, but the populations do not usually suffer noticeably losses during winter (OOSTENBRINK 1966). Laboratory research showed that -16° C kills *Anguina tritici* in infected wheat seeds. SAYRE (1963) reported that the eggs of some nematode species tolerated - 35°C for a considerable time. Other examples of nematode tolerance to low temperatures are reviewed by DAO (1970). Freezing as a technique for nematode control has hardly been applied in practice.

#### 2.1.3. Drought

Fallow as a practice to increase soil fertility was known of old. The ancient Egyptian 'sheraqi' induced temperatures of over 46°C in the top ten centimeters of soil in middle- and upper-Egypt (PRESCOTT 1919 and 1920). Several authors have mentioned the possible value of this method for nematode control

(CRITTENDEN 1953, CHRISTIE 1959, RASKI 1959, SEINHORST 1968 and KHAN pers. comm.).

#### 2.1.4. Flooding

BROWN (1934) reported that flooding the soil for 12-22 months would be necessary to obtain a good control of root-knot nematodes. CRALLEY (1957) found that flooding of rice fields at sowing time reduced white tip disease, caused by *Aphelenchoides besseyi*, from 60% to 1%. JOHNSON (1957) stated that survival of *Tylenchorhynchus martini* was poor in rice fields saturated with water. HOLLIS and RODRIGUEZ (1966) found the nematicidal compounds butyric acid and propionic acid in the soil shortly after flooding. Usually flooding has to be continued for several months to make it effective against nematodes. The technique is therefore not very promising, unless special crops can be grown in the wet soil as a rotation practice, like rice and reed.

#### 2.1.5. Electricity

DAULTON and STOKES (1952) reported excellent root-knot control by passing an electric current through soil, but LEAR and JACOB (1955) got negative results. Electricity is, of course, effective as a source of heat as recorded under 2.1.1

#### 2.1.6. Irradiation

Radioactive irradiation was studied as a method of nematode control (MYERS 1960, GREEN and WEBSTER 1965), but it was not effective in economic doses for sterilization of soil (WOOD and GOODEY 1957) nor applicable for killing nematodes in plants (MYERS and DROPKIN 1959, VAN DE WOESTIJNE and VAN DEN BRANDE 1960). Other kinds of irradiation also had little effect on nematodes, e.g. ultraviolet irradiation (GREEN and WEBSTER 1965) and ultrasonic waves (KÄMPFE 1962).

#### 2.1.7. Mechanical disturbance

Ploughing, harrowing and rotavating soil may kill or inactivate part of the nematofauna, although the effect of these methods is usually small unless they are combined with fallow. CHRISTIE (1959) recommended ploughing immediately after harvest to expose infested roots to heat and drought. OOSTENBRINK (1964) recorded effective suppression of *Trichodorus teres* by rotavating the soil. Beneficial effects obtained by deep ploughing or by burying the top soil are recorded, but they are rather soil replacement than PSS and fall outside the scope of this publication.

## 2.2. CHEMICAL MEANS

#### 2.2.1. Early nematicides

These chemicals comprise carbon bisulfide, hydrocyanic gas, cyanamides, isothiocyanates, hypochlorites and chloroacetates. KÜHN (1881) attempted to

eradicate *Heterodera schachtii* by means of carbon bisulfide, known from applications against *Phylloxera* infestation of grape in France. BYARS (1919) was able to control root-knot nematodes by means of hydrocyanic gas. Both researchers reported that the partial soil sterilization obtained was not economic. The herbicide sodium cyanamide was tested by WATSON (1924). MILES and TURNER (1928) reported on the application of calcium and sodium cyanamide against root-knot in tomato and cucumber. HURST and FRANKLIN (1938) evaluated calcium and sodium cyanamide against *H. rostochiensis*, TRIFFITT (1929) did the same with mustard oil (isothiocyanate), SMEDLEY (1936 and 1939) with hypochlorite and isothiocyanates, SMEDLEY (1938) and O'BRIEN et al. (1939) with chloroacetates. Of these chemicals only calcium cyanamide is still studied in connection with certain special applications, whereas we will meet isothiocyanates again as the active ingredient in some modern nematicides.

The number of chemicals used as soil disinfectants is still small. Nematicides account for most of the treated area. Application of nematicides on a practical scale came to the fore only after the discovery of the halogenated hydrocarbons DD (CARTER 1943) and EDB (CHRISTIE 1945) as relatively cheap, effective chemicals. Some nematicidal chemicals were known earlier, as well as the broad-spectrum soil fumigants chloropicrin and methyl bromide. Several others, belonging to different chemical groups, have been discovered since then. A number of specific soil fungicides are also known. The most important chemicals used for PSS are briefly discussed under the headings: broad-spectrum soil fumigants, halogenated hydrocarbons, isothiocyanates, organophosphorous compounds and other systemics, soil fungicides, other soil pesticides. The chemicals used frequently in our experiments are listed in Table 1, which indicates their chemical structure and other important data.

#### 2.2.2. Broad-spectrum soil fumigants

The main soil fumigants which are effective against all living organisms in the soil, are chloropicrin and methyl bromide.

##### a. Chloropicrin (CP)

The first report concerning CP as an effective nematicide was by MATTHEWS (1919); it was known as an insecticide and previously as a war gas; GODFREY (1934 a & b) applied CP against the root-knot nematode in Hawaii and got better yields and quality. TAYLOR and MCBETH (1941b) introduced planting-site treatment for melon. Good results against root-knot nematodes were reported by NELLER and ALLISON (1935), CHITWOOD (1941), TAYLOR (1943), STARK et al. (1944), TAYLOR and MCBETH (1947), MACHMER (1949) and DARBY (1961) and others. CHITWOOD and NEWHALL (1942) were able to control onion bloat caused by stem eelworm in muck soils. Destruction of *Pratylenchus* species was reported by OWEN and ELLIS (1951), RASKI (1956), LEMBRIGHT (1959) and APT and GOULD (1961). BAINES and MARTIN (1953) recorded control of the citrus nematode *Tylenchulus semipenetrans* down to a depth of five feet. DARBY (1961) and others studied the effect of CP against ectoparasitic root-infesting nematodes,

viz. *Trichodorus christiei*, *Belonolaimus longicaudatus*, *Hemicycliophora parvana*, *Hoplolaimus tylenchiformis* and other species.

Successful control of soil-borne pathogenic fungi by CP was reported by ANONYMOUS (1950a & b), WILHELM (1965), LEMBRIGHT (1959), RASKI (1956), CONVERSE (1960) and many others. HOESTRA (1968) introduced CP application as a practical method for controlling apple replant disease. APT and GOULD (1961) reported slight increase in losses from narcissus *Fusarium* basal rot due to CP soil treatment in spite of yield increase over check and top grade. BUCHENAU (1963) observed better emergence, winter survival, plant length, yield and kernel weight for wheat, when the soil was treated with CP. This treatment of the soil also caused better yields and quality for pine-apple (JOHNSON and GODFREY 1932; TAM and CLARK 1943 and TAM 1945), peach (TAYLOR and MCBETH 1947), several fruits (MACHMER 1949), orange (BAINES and MARTIN 1953), tobacco (TAYLOR 1956), strawberry (LEMBRIGHT 1959); narcissus (APT and GOULD 1961) and tomato (STARK et al. 1944). Sealing of CP treated soil is important to obtain good results (TAYLOR 1943) and to save part of the dose (LEMBRIGHT 1959). The effect of a CP-treatment of the soil may last for two (OWENS and ELLIS 1951) or even four years (LEMBRIGHT 1959) and sometimes the yield is still favourably influenced 5 years after the treatment (MACHMER 1949 and BAINES and MARTIN 1953). In some cases, however, soil-borne diseases returned more seriously some years after a CP treatment than in the control soil (OOSTENBRINK and HOESTRA 1961, APT and GOULD 1961).

Weed control may also be an important result of CP treatment (TAYLOR 1956 and DARBY 1961). PARRIS (1958) reported that CP is capable of destroying protoplasm in any organism and therefore emphasized the power of CP to kill all kinds of soil organisms.

#### *b. Methyl bromide (MB)*

RICHARDSON and JOHNSON (1935) mentioned MB as an effective nematicide for high value crops. TAYLOR and MCBETH (1940, 1941 a & b) emphasized the efficacy of MB against many kinds of soil organisms and indicated the necessity of sealing the treated soil to obtain good results. Root-knot nematodes were reported to be suppressed by MB soil treatments (SHER et al. 1958, MORGAN 1958, THOMASON 1959, DARBY 1961 and GOOD 1964), as well as *Pratylenchus* species (OAKES et al. 1956, APT and GOULD 1961 and RADEWALD et al. 1964), *Trichodorus christiei* and related species (OAKES et al. 1956, DARBY et al. 1962, ADAMS and TRUE 1962 and RADEWALD et al. 1964), *Tylenchorhynchus martini*, *T. dubius* and other species (OAKES et al. 1956, ATKINS and FIELDING 1956, ADAMS and TRUE 1962), *Belonolaimus gracilis* and *B. longicaudatus* (STEEL and GOOD 1958, DARBY et al. 1962), *Hoplolaimus tylenchiformis* and *H. galeatus* (DARBY et al. 1962, RUEHLE and SASSER 1964), *Hemicycliophora parvana* (DARBY et al. 1962), *Meloidodera floridensis*, *Helicotylenchus dihystrera* and *Xiphinema americanum* (RUEHLE and SASSER 1964), and cyst forming nematodes (HAGUE and SOOD 1963). CHEN et al. (1962) reported good nematode control for a period of 3 years, when MB was applied.



The effect of MB on soil fungi is apparently less marked than that of CP (STARK and LEAR 1947). It did not control some *Fusarium* species (McCLELLAN et al. 1947, APT and GOULD 1961), but was effective against others (THOMASON 1959, GOOD 1964). McCLELLAN et al. (1947) reported that MB was moderately effective against *Sclerotium rolfsii*. THOMASON (1959) found it effective against *Sclerotia bataticola*, but not against *Rhizoctonia solani* and *Stemphyllium* sp. It reduced *Phytophthora fragaria* infestation of strawberry (CONVERSE 1960), and was effective against damping off problems caused by *Pythium* spp., *Pellicularia filamentosa* and *P. rolfsii* (DARBY 1961).

Crop yields and quality were often reported to be increased by MB treatment without specifying the cause. Such reports were encountered on tomato (STARK et al. 1944), tobacco (TAYLOR 1956, MORGAN 1958), rice, corn and Lima beans (ATKINS and FIELDING 1956, OAKES et al. 1956, STEEL and GOOD 1958), narcissus (APT and GOULD 1961), Norway spruce and pine (ADAMS and TRUE 1962, RUEHLE and SASSER 1964). Alfalfa also showed increased yields for the second year after treatment (RADEWALD et al. 1964), and Norway spruce continued to show better growth for 5 years (ADAMS and TRUE 1962).

Records concerning herbicidal effects of MB were reported by TAYLOR (1956), DARBY (1961), GOOD (1964) and others. MB affects the protoplasm of nematodes and other animals, fungi and weeds, according to PARRIS (1958).

### 2.2.3. Halogenated hydrocarbons

This group comprises the three most successful nematicides as well as some other effective chemicals, widely used as soil disinfectants, viz. dichloropropene, ethylene dibromide and dibromo-chloropropane. Methyl bromide also belonging to this group, has already been discussed under (2.2.2.b).

#### a. Dichloropropene (D)

This is the active ingredient of DD (a mixture of more than 50% dichloropropene with dichloropropane and some other compounds), also available without secondary compounds under other names.

CARTER's 1943 publication on DD is classic now, because it introduced the first nematicidal soil fumigant which became successful from a technical and an economical point of view. He reported that DD was effective against soil animals but not against fungi in standard doses. Since then numerous reports on DD or other formulations of dichloropropene have appeared. Early reports showed it to be successful against root-knot nematodes in watermelon (PARRIS 1946), peach (TAYLOR and MCBETH 1947), tobacco (ANDERSON 1951, SHEPHERD 1952, MILLER 1957), cotton (RASKI and ALLEN 1953, YOUNG and SMITH 1953), sweet potato (MULLIN 1952, NIELSEN and SASSER 1959), tomato (MILLER 1958, MARLATT and ALLEN 1959), sugarbeet (MILLER 1958), grapes (RASKI and LEAR 1962), strawberries (KANTZES and MORGAN 1962), caladiums (RHOADES 1964), onion (THOMASON et al. 1964) and other crops (CHAMBERS and REED 1963, WILSON and HEDDEN 1964, and several others). DD also controls cysteelworms, e.g. the beet cyst nematode (THORNE and JENSEN 1946) and the

potato cyst nematode (PETERS 1948, SCHMITT 1948, OOSTENBRINK 1950 and others).

Successful control of *Pratylenchus* species has, among others, been obtained in tobacco (ANDERSON 1951), corn (OWENS and ELLIS 1951, OAKES et al. 1956, YOUNG 1964, EDMUNDS et al. 1967), nursery stock (HOESTRA 1961, OOSTENBRINK 1961, OOSTENBRINK and HOESTRA 1961 and HOESTRA and OOSTENBRINK 1962), walnut (LOWNSBERY and SHER 1957), onion (THOMASON et al. 1964), narcissus (APT and GOULD 1961, SLOOTWEG 1954). DD was also effective against *Rotylenchulus reniformis* in cotton (ANDERSON 1956), *Tylenchulus semipenetrans* in citrus (BAINES and MARTIN 1953, BAINES et al. 1962), *Ditylenchus* species in daffodil (THOMAS et al. 1958), potato (DARLING 1959) and onion (BIRD and SMITH 1965), *Trichodorus* species in onion (THOMASON et al. 1964, JENSEN et al. 1964), *Tylenchorhynchus* species in corn (OAKES et al. 1956), rice (ATKINS and FIELDING 1956, HOLLIS et al. 1959) and Norway spruce (ADAMS and TRUE 1962), and also *Belonolaimus gracilis* (STEELE and GOOD 1958, YOUNG 1964), *Xiphinema americanum* (THORNE and SCHIEBER 1962), *Criconeoides* species (YOUNG 1964) and *Pratylenchus* species (WILSON and HEDDEN 1964). Dichloropropene is effective against all soil animals, including soil insects (CHRISTIE 1947a & b), although it is hardly used against animals other than nematodes for economic reasons.

The effect of dichloropropene on soil fungi was studied by a number of authors. MCCLELLAN et al. (1947) and others, found that DD was not effective against fungi. It is not recommended for the control of soil-borne fungus infestations. Several authors, however, indicate that it has fungicidal effects when the dose is considerably increased (WENSLEY 1956). YOUNG and SMITH (1953) found that DD was effective against *Fusarium* wilt of cotton; GOOD (1964) found the same for *Fusarium* infestation of okra. BAINES et al. (1962) reported that high doses of DD controlled *Fusarium* and *Pythium* species in soil treatment prior to planting lemon, and HOESTRA (1968) recorded strong effect of high doses of DD for the removal of replant disease of apple in pot experiments.

Increased plant growth and quality due to dichloropropene treatment was reported by TAM (1945) for pineapple, PARRIS (1946) for watermelon, TAYLOR and MCBETH (1947) for peach, PETERS (1948), SCHMITT (1948) and PETERS (1949) for potato, ANDERSON (1951), MCBETH (1951) and MILLER (1957) for tobacco, MCBETH (1951) and MILLER (1958) for tomato, MCBETH (1951) for melon and cowpea, MULLIN (1952) and NIELSEN and SASSER (1959) for sweet potato, ALLEN and RASKI (1952) for tuberous *Begonia*, OAKES et al. (1956) and YOUNG (1964) for corn, THOMASON et al. (1964) and JENSEN et al. (1964) for onion, LOWNSBERY and SHER (1957) for walnut, MILLER (1958) for sugar beet, HOLLIS et al. (1959) for rice, ADAMS and TRUE (1962) for Norway spruce, BAINES and MARTIN (1953) for orange, BAINES et al. (1962) for lemon and RHOADES (1964) for *Caladium* tubers. The positive effects of a DD treatment may be evident for one year (MCBETH 1951), two years (ANDERSON 1951, LOWNSBERY and SHER 1957) even up to ten years (BAINES et al. 1962), depending on the problem and the dose used.

Several workers report negative effects of dichloropropene treatments due to phytotoxicity, which may reduce plant growth and influence taste of the product (BESEMER and OOSTENBRINK 1955).

MARLATT and ALLEN (1959) got unsatisfactory tomato yields, when DD was used to control root-knot nematodes due to a heavy, unexpected virus disease. ANONYMOUS (1950a&b) and SHEPHERD (1952) reported reduced quality of tobacco.

ENNIK et al. (1964) found that DD reduced clover in a grass-clover mixture, whereas KOOISTRA (1964) concluded that DD increased the ability of white clover to compete with perennial ryegrass.

GOOD and RANKIN (1964) reported that DD caused a moderate weed control effect, which observation was also made by other workers.

#### *b. Ethylene dibromide (EDB)*

CHRISTIE (1945) was the first worker to indicate the efficacy of EDB as a nematicide. Effective nematode control and yield increase were also reported by: NEWHALL and LEAR (1948), TARJAN (1948), ANDERSON (1951), SHEPHERD (1952), BAINES and MARTIN (1953), YOUNG and SMITH (1953), ATKINS and FIELDING (1956), HORN et al. (1956), WILSON (1956), LOWNSBERY and SHER (1957), MILLER (1957), STEEL and GOOD (1958), THOMAS et al. (1958), MARLATT and ALLEN (1959), NIELSEN and SASSER (1959), CHAMBERS and REED (1963) and WILSON and HEDDEN (1964).

The effect of EDB was amongst others, studied in citrus, ornamentals, tobacco, strawberry, rice, and vegetable crops and for control of citrus, root-knot, stem, stilet, lesion, sting and burrowing nematodes, as well as wilting and complex diseases, in various crops.

PEACHEY and WINSLOW (1962) reported that EDB caused a partial sterilizing effect in soil.

MCCLELLAN et al. (1955) noticed no decrease of wilting in cotton after the application of EDB. MORGAN (1957), REYNOLDS and HANSON (1957) and DICKEY (1962), however, noticed that EDB influenced wilting of tobacco, damage by *Rhizoctonia solani* to cotton and artificial bacterial infestation on tomato.

#### *c. Dibromochloropropane (DBCP)*

MCBETH (1954) and RASKI (1954) published the first indication that DBCP, in the formulation Nemagon, was a valuable nematicide. Due to its low vapour pressure, it is effective only in warm soils, therefore in glasshouses, and in tropical and sub-tropical regions. Good results with DBCP were obtained against the citrus nematode (O'BANNON and REYNOLDS, 1962 and 1963), against root-knot nematodes in tobacco (MORGAN 1958), cotton (MORTON 1959), strawberry (POTTER and MORGAN 1956, MORGAN and JEFFERS 1957, SMART et al. 1967) and carrot (WILSON and HEDDEN 1964), against *Ditylenchus dipsaci* in soil (BARKER and SASSER 1959), against *Belonolaimus longicaudatus* and *B. gracilis* (COOPER et al. 1959, STEEL and GOOD 1958), against *Trichodorus* species (MINTON et al. 1960), against *Radopholus similis* in citrus (SUIT et al.

1961). LEAR et al. (1965) reported good control of *Heterodera schachtii* and *H. cruciferae*, whereas control of *H. schachtii* was poor in experiments by MILLER (1958). Poor results with DBCP were also reported for root-knot nematodes in tomato by MARLATT and ALLEN (1959), and for *Pratylenchus* species in walnut by LOWNSBERY and SHER (1957).

DBCP is used in practice against nematode infestations only, although some effect against soil fungi and bacteria has been observed, e.g. against *Rhizoctonia solani* in tomato (ASHWORTH et al. 1964), against *Sclerotium rolfsii* in Texas spinach and peanuts (THAMES and LANGLEY 1964) and against artificial infestation of *Agrobacterium tumefaciens* (DICKEY 1962).

BIRCHFIELD and PINCKARD (1964) and BIRD (1963) were able to control *Fusarium* wilt and root-knot nematodes in cotton simultaneously by using a mixture of DBCP and the fungicide pentachloro-nitrobenzene.

DBCP is successful in practice, because it is effective at small doses and therefore at a reasonable price, and because phytotoxicity is so low that many crops can be treated during growth without suffering noticeable damage.

#### *d. Other halogenated hydrocarbons*

Several other halogenated hydrocarbons are effective nematicides, e.g. dichloroethane, chlorobromopropene, dichlorophenylpropene and a number of related compounds. They did not find wide application, but good results were obtained by Russian workers (ČEKALINA 1966, VOLODKOVIČ and ČEKALINA 1967) in experiments against root-knot nematodes and beet cyst nematodes. Some of these chemicals possess fungicidal qualities.

#### *2.2.4. Methylisothiocyanate compounds (Trapex and Vapam)*

Methylisothiocyanate (MIT) was already recorded as an effective nematicide by both TRIFFITT (1929) and SMEDLEY (1936, 1939), and it later appeared to have some antibiotic qualities against other animals, fungi, bacteria and weeds as well. Two modern nematicides based on MIT are Trapex and Vapam. Trapex is a 20% formulation of MIT, which was introduced by PIEROH, WERRES and RASCHKE (1959); Vorlex is a mixture of Trapex with dichloropropene formulations. Vapam, recorded as a nematicide by LEAR in 1956, comprises 20–40% Sodium-N-methyldithiocyanate (metham sodium) which in the soil is transformed to MIT within a few hours after application. The chemical is also effective when it is introduced as a solution in irrigation water and can also be used in hydroponic cultures (SVEŠNIKOVA 1965). Trapex and Vapam are comparable in their effect; both are strongly phytotoxic and require a waiting period of several weeks after treatment before plants can be sown or planted (CHANDRA and BOLLEN 1961). Both chemicals are used in practice on a limited scale against specific problems in which pathogens other than nematodes are involved.

Positive results with Trapex or Vapam were obtained against nematodes (ADAMS and TRUE 1962, GOOD and RANKIN 1964, and others), nematodes and fungi (BAINES et al. 1957 and 1958), *Streptomyces scabies* (FINK 1956), bacteria (DICKEY 1962) and unknown causes of yield deficiencies (PEACHEY and WINSLOW 1962).

### 2.2.5. Organophosphorous compounds and other systemics

Several insecticides, such as DDT, BHC and dieldrin, are not, or only slightly effective against nematodes, but many organophosphorous compounds are. Most of them are not very phytotoxic, work as systemics and can be applied as sprays or dips for plants and plant organs or as soil sterilizers. The list of effective chemicals comprises parathion, systox, malathion, disyston, diazinon, thimet, nellite, V.C. 13, fensulfothion, thionazin, lannate and others. Early reports on the effect of parathion and systox sprays against leaf eelworms and stem eelworms in chrysanthemum, strawberry, alfalfa and other crops are by RASKI and ALLEN (1948), DIMOCK and FORD (1950), BROWN and FRANKLIN 1953, FEDER (1952, 1954), BERGESON (1955) and others. These chemicals are also effective against root eelworms when they are mixed through the soil.

The two compounds last mentioned are among the promising systemic nematocides, of which a number of formulations are under investigation, such as Terracur P and Dasanit on the basis of fensulfothion = 0.0-diethyl-0-(4-methylsulphinyl-phenyl)-monothiophosphate, and Nemafofos, Zinophos and Cynem on the basis of thionazin = 0.0-diethyl 0-2-pyrazinyl phosphorothioate.

Other new promising nematocides outside the group of the organophosphorous compounds comprise Temik, on the basis of aldicarb = 2-methyl-2-(methylthio) propionaldehyde-0-(methylcarbamoyl)-oxim, and a number of experimental chemicals.

Due to toxic effects of residues, it is uncertain whether any member of these systemic nematocides could be released for application on food crops.

### 2.2.6. Soil fungicides

Two fungicides, which were often used in our study, are Captan and PCNB. They are known as fungicides, but preliminary experiments showed that each of them killed about half of the plant parasitic nematodes when applied to soil in the usual amount. Their nematocidal effect, therefore, is not negligible.

#### a. Captan (CA)

The active ingredient of Captan is N-(trichloromethylthio)-1,2,3,6tetra-hydro-phthalimide. It was introduced as a fungicide in 1949 by the Standard Oil Development Co. KITTLESON (1952) was the first to describe the compound as an effective fungicide particularly for application to foliage. In the soil Captan has a half life of 70-75 days. It significantly reduces the total number of fungi, bacteria and actinomycetes. Among the fungi effectively controlled were *Pythium ultimum* and *Rhizoctonia solani* (DOMSCH 1959). HOESTRA (1968) investigated the effect of Captan treatment in connection with research on a specific apple replant disease in The Netherlands. In pot experiments some stimulation of growth in apple was observed. In the field the effect was only slight. DOMSCH (1959) and SPENCER (1968) reported no phytotoxic action of this chemical, while EISSA (1971) observed phytotoxicity when working with barley.

#### *b. Pentachloronitrobenzene (PCNB)*

The fungicidal and bactericidal effect of PCNB when applied as a soil sterilizer is reported by many workers (Auctores diversi). RANKIN and GOOD (1959) and RANKIN (1961) described a phytotoxic effect of this compound. MURANT and TAYLOR (1965) indicated that PCNB also suppressed nematode populations effectively, in this case of *Longidorus elongatus*. MILLER (1961) and SUIT et al. (1961) reported that PCNB mixed with Nemagon increased apple and citrus growth, and BIRD (1963), and BIRCHFIELD and PINCKARD (1964) found the same with regard to cotton. MILLER and WAGGONER (1962) found that PCNB controlled soil pathogenic fungi but doubled the nematode population because its natural enemies were suppressed.

#### *2.2.7. Other soil pesticides*

Several other chemicals are known to be powerful soil disinfectants, but have for some reason or other not yet reached widespread application. Most of them are nematicides; at least some of them are effective against a wide range of organisms. The list comprises amongst others: sodium-selenate, a powerful systemic nematicide and insecticide when applied to the soil, according to DIMOCK (1944) and TARJAN (1950a); copper-sulfate, which may kill nematodes in doses which are not yet phytotoxic (DE MAESENEER 1967);

mercury compounds, namely chlorides and oxide, which have to be mixed through the soil and are effective against nematodes without causing toxicity of the plant and the soil, according to GRAINGER (1956, 1960) and GRAINGER and CLARK (1963); several sulfur-compounds, such as N, N' dimethylthiuram-disulfide (Tridipam), tetrachlorothiophene and dichlorotetrahydrothiophene (BOOGAERT and HIJINK 1959, MILLER and WAGGONER 1962);

sugar, when applied at high doses (FEDER, EICHHORN and HUTCHINS 1962); dinitroorthocresol, which is a wellknown herbicide and insecticide, but is also applied to de-nematize mushroom sheds in The Netherlands, and sometimes as soil disinfectant against nematodes in East-European countries (STOJANOV 1962);

formaldehyde, which is effective against fungi and nematodes when applied in water at doses of 15-20 l/m<sup>2</sup>;

calciumformate and other fatty acid derivates (VAN BERKUM 1964, JOHNSTON 1957); calciumoxide, viz. burning lime, which is effective if applied at a high dose to soils with a high pH (KUIPER and DE LEEUW 1963).

The normal fertilizers may also affect the density of plant nematodes and possibly of other pests and pathogens in the soil when a heavy dosage is applied (OOSTENBRINK, pers. comm.).

### **2.3. PSS EFFECTS**

PSS causes direct effects, in relation to the added materials, the organisms in the soil and the soil itself, and indirect effects, e.g. on plant growth. Plant growth

for practical reasons was used as a yardstick to measure the effects of PSS in this study. The relation of plant reactions to the direct effects of PSS, therefore being an interpretation of these reactions, was not the main purpose of this study. It was nevertheless pursued in a number of experiments. For this reason the present knowledge of the influence of PSS on plant growth-factors was reviewed under the heading: elimination of noxious organisms, effect on soil fertility, and direct effect of soil sterilizers.

### *2.3.1. Elimination of noxious organisms*

This is the principal incentive for the application of PSS. Certain treatments destroy all organisms in soil, and others are selective, as indicated before. The main groups of organisms controlled by PSS are: nematodes and other animals, fungi and bacteria, and complex diseases (including soil borne viruses with nematodes or fungi as vectors).

#### *a. Nematodes and other animals*

Most nematicides as well as the broad spectrum treatments, without much selectivity, kill all species of nematodes except the saprozoic nematodes which usually survive better. Some differences may occur in toxicity to various stages in the life cycle of a given species and to different species (ANONYMOUS 1968), but these differences are small. The killing effect largely depends on the chemical used, the dose, method of application, type and structure and also moisture and temperature conditions of the soil, and whether the nematodes are protected in parts of roots, cysts or otherwise. It therefore depends on many variable factors and a 100% kill is hardly ever attained.

Little is known about the mechanisms of nematode kill by different sterilizers, although stray bits of information and suggestion have been published (CHITWOOD 1952, RHODE 1960, WELLE and BIJLOO 1965, BROWN and DUNN 1965, KRUSBERG 1965, 1968, SPURR 1966, MARKS et al. 1966, ANONYMOUS 1968). Thermal kill, possibly due to protein and fat denaturation, has been discussed by MORITA and GODFREY (1933), BELEHRADEK (1935), STANILAND (1953), LEVITT (1956), GIESE (1957), KOFFLER et al. (1957), WALKER (1960 & 1962), BLOOM (1963) and SAYRE (1963).

Nematicides are usually zoocides and kill all animals in soil, and in the case of being systemic also in leaves. They are therefore effective, and sometimes used against wireworms or other soil insects, and against aphids (LANGE 1947, RHOADES 1961, MOTSINGER 1961, and others).

#### *b. Fungi and bacteria*

Fungi vary much in their susceptibility to the means used for PSS. An important point still being discussed is whether the widely used nematicides, DD and EDB have little effect against soil fungi or none at all (PARRIS 1945, CHRISTIE 1947a & b and others). Other workers, however, reported some effect, at least against certain fungi, of DD and EDB (ZENTMYER and KENDRICK 1949, JACK and SMITH 1952, and others), especially with high doses of the chemicals (HOESTRA 1968).

CP, MB and other broad-spectrum disinfectants as well as DBCP and Captan are reported to be effective against many soil fungi by numerous workers. In some cases PSS finally led to an increase of a fungus infestation after an initial suppression, e.g. for *Sclerotium rolfsii* on tomato (RANKIN and GOOD 1959), or an otherwise harmless species became a noxious parasite after PSS, e.g. *Trichoderma viride* on alfalfa and celery (EDMUNDS and MAI 1966). The results of PSS against pathogenic bacteria are not well known and are often contradictory. Heat and CP are known to be effective in most cases, but a number of reports indicate that DD also has a noticeable effect against bacteria, e.g. *Bacterium solanacearum* in tomato (SMITH 1947) and *Pseudomonas syringae* in peach (DEVAY et al. 1962).

### *c. Complex diseases and soil-borne viruses*

Soil-borne plant diseases are often of a complex character and nematodes are quite frequently members of such complexes according to STEINER (1953). Many nematode-fungus, nematode-bacterium, and nematode-virus complexes have been identified in recent years, as well as some fungus-virus complexes (PITCHER 1965, and CALVERT and HARRISON 1966).

Complex diseases can usually be cured by the elimination of one of the constituents and this makes application of nematicides effective against problems in which nematodes are involved as principal pathogens, initiators, contributors or vectors.

### *2.3.2. Effect on soil fertility*

PSS influences soil fertility by changing the availability of nitrogen, but its influence on other plant nutrients and on physical soil properties may also be noticeable.

PSS usually causes a nitrogen flush, because part of the animal and plant organisms in soil are destroyed and mineralized (ammonified); it may also enhance the ammonium/nitrate ratio in the soil, owing to the new ammonium formed and to the fact that nitrification in treated soils is temporarily reduced (DU BUISSON 1917, STARK et al. 1939, KINCAID and VOLK 1949, WINFREE and COX 1958, CHANDRA and BOLLEN 1961, GORING 1962, ALTMAN 1963, EDMUNDS et al. 1967 and HIJINK 1969).

The increase of available nitrogen was estimated to be some tens of kilogrammes per ha usually (OOSTENBRINK 1957). TILLET (1964) mentions 10–30 kg/ha for sand and silt soils in Rhodesia. KOLENBRANDER (1969), however, indicated that the amount increased with the humus content of the soil; he also concluded that autumn application of DD in The Netherlands did not cause a strong nitrogen flush, but nevertheless markedly increased the nitrogen availability in the spring because it suppressed nitrification and therefore loss of nitrogen during the winter, since nitrate is drained away in a rainy climate whereas ammonium is not. Delayed nitrification was therefore considered more important than the direct nitrogen flush itself. When rapid nitrification occurs, the nitrogen available in the soil may soon be equal or less for treated as com-



pared to untreated soil. Application of DD in Hawai stopped nitrification for 8 weeks, whereas the influence of CP was still measurable after 2 years (TAM 1945). COLBRAN and GREEN (1963) found a delay of 60–110 days after application of DD, CHANDRA and BOLLEN (1961) recorded 30–60 days for Nabam and Mylone. ALTMAN (1963) found an excess of ammonium for 7 weeks after treatment with DD. High temperature, high organic matter content, re-inoculation and a high pH promoted quick restoration of the nitrification process (GORING 1962). The high amount of ammonium-nitrogen in treated soils may be favourable to the crop, e.g. for pineapple (TAM 1945) and for potato (PETERS 1953), or it may be harmful, e.g. for tobacco (TILLET 1964) and for tomato, bushbean and squash (GOOD and CARTER 1965). The last mentioned authors recommended addition of extra nitrogen fertilizer shortly after PSS.

Other plant nutrients may also be influenced. Nematode infested plants often comprise a lower content of certain minerals than uninfested (OTEIFA 1952, SHER 1957a & b). Mineral deficiencies may be indicative of nematode infestation (RASKI 1953). PSS, either chemical or thermal, may influence the balance of minerals in the soil, and therefore the mineral contents of plants. On treated soil the leaves of boxwood comprised more Na (TARJAN 1950b), the leaves of lemon comprised more Ca, Mg and less Na and the leaves of orange comprised more K and Mn (MARTIN et al. 1953). Heating of the soil may cause excess of Mn which may cause manganese intoxication in lettuce (MESSING 1965). SONNEVELD (1968) reported that release of Mn depends on heating temperature, time of exposure, and soil type. Incidentally PSS with CP and MB caused zinc deficiency in cotton (WILHELM et al. 1967). EDMUNDS et al. (1967) found more Fe and Al and less Mn and nitrate in maize leaves after PSS with either heat or chemicals. The effect of PSS may differ with soil type and plants and perhaps other factors. Further, uncertainty exists in many cases whether differences in plant composition are due either to differences in mineral composition of the treated as compared to the untreated soil, or to better root development due to elimination of noxious organisms.

A slight increase of the soil pH following PSS was reported by OOSTENBRINK (1958). This might have been due to the ammonium accumulation recorded earlier, for the change of ammonium into nitrate again suppresses the pH (CHEVRES-ROMAN 1967).

### 2.3.3. *Direct influence of soil pesticides on plants*

Direct growth-promoting effects of soil pesticides on plant growth are possible if they include nutrient elements, but may be discarded in the case of fumigants which disappear rapidly. The amounts are usually small in term of fertilizer application. Nevertheless some positive effects have been reported. ALTMAN and LAWLOR (1964) reported that certain bacteria feed on DD as a C-source and produce plant-growth promoting substances.

However, direct effects of soil pesticides are usually noxious. Most soil disinfectants are strongly phytotoxic and application must therefore be followed by a waiting period from one to several weeks before sowing or planting is

possible. Also when plant growth is increased by the treatment, phytotoxicity may be involved and may have decreased the effect of the treatment. Sometimes the cause of phytotoxicity is specifically related to a certain component of the pesticide, as for instance yield reduction of cabbage, bean and beet after MB and EDB treatments which is attributed to a high bromine-content (STELMACH 1959). A number of soil pesticides may influence quality and taste of harvested products, or cause toxic residues in such products (BESEMER and OOSTENBRINK 1955).

### 3. MATERIALS AND METHODS

#### 3.1. INVENTORY OF DATA ON PSS EFFECTS

More than 2400 items on the effect of a soil disinfection on a certain plant in a certain soil in The Netherlands could be traced and were incorporated in a file of computer cards for statistical treatment. The data were collected from publications, and from unpublished files of the Plantenziektenkundige Dienst (PD), the Landbouwhogeschool (LH), working parties of the Nationale Raad voor Landbouwkundig Onderzoek (TNO), and individual workers, and were complemented with 545 items from the author's own field and pot experiments.

#### 3.2. FIELD EXPERIMENTS, GLASSHOUSE AND LABORATORY STUDIES

A standardized series of field trials in locations all over The Netherlands was treated in 4 different ways and cultivated with 7-14 crops in 1967. A general biocide, a nematicide and a fungicide in light to moderate doses were applied in comparison with control. The trials were realized in cooperation with the Agricultural Department of the PD and its local offices. Soil from a number of these fields was also extensively studied in glasshouse and laboratory experiments.

Other glasshouse experiments were made to investigate response to PSS for different types of soil and for soils in which a specific pest or disease was present, and to study the effect of inoculations.

#### 3.3. NEMATICIDES AND THEIR APPLICATION

A hand injector was used for field application of most fumigants. Methylbromide was applied directly from the original container under plastic cover because of its high vapour pressure. Chloropicrin was also applied under plastic cover. For treatment of pots with soil the chemicals were pipetted near the temporarily sealed pot bottom; in other experiments the entire soil quantity was treated in a plastic bag placed inside a sealed metal bin. Usually one week after treatment aeration was started and maintained for 3 weeks; garden cress (*Lepidium sativum* L.) was sometimes used to check whether phytotoxic residues were still present. Granular and powdery chemicals were mostly mixed with the soil by means of hand tools or a rototiller.

### 3.4. TEST PLANTS AND THEIR EVALUATION

In most cases seeds of local Dutch cultivars of the crops tested were used. The commonly used criteria to evaluate plant growth were: germination, length, thickness, fresh and dry weights of roots or total plants or harvested parts of the plants. Visual evaluation figures were also used to rate plant vigour, size and color as well as numbers of plants, leaves, ears, seeds, flowers or fruits.

### 3.5. NEMATODES AND THEIR ENUMERATION

Soil samples were extracted and analysed by OOSTENBRINK's technique (1960). Root-lesion nematodes were extracted from roots by the blending-cottonwool technique (STEMERDING 1963). For root-knot nematodes, incubation in a funnel-spray apparatus was used (OOSTENBRINK 1960), which combines BAERMANN's funnel (1917) and a mistifier (SEINHORST 1950). A visual index was used to indicate the infection rate for root-knot nematodes, in which the rates 0, 1, 2, 3, 4 and 5 indicated that 0%, 1-10%, 11-20%, 21-30%, 31-50% and 51-100% of the total root system showed galls. A similar index was sometimes used to classify infestation by cyst-forming nematodes.

### 3.6. STATISTICAL TREATMENT OF THE DATA

The experimental results were usually tested by analysis of variance and other significance tests, as indicated in the tables (SNEDECOR 1956). Least significant differences were often calculated.

The IBM and CDC computer facilities of the Landbouwhogeschool Computer Centre were used to perform most of the statistical tests. The standard programmes (437 K) and (441 La) were used, as well as a special programme (Keuls pers. comm.) and a number of other specific tests and data transformations.

All treatments which resulted in a yield lower than that of the untreated control were classified as 'toxic cases'.

## 4. INVENTORY OF QUANTITATIVE DATA

### 4.1. INTRODUCTION

Each field or pot experiment in which a test plant is grown in disinfested as compared to untreated soil, resulted in a figure indicating the effect of a treatment on the growth of a plant. Expressed as a percentage of untreated, it may vary from infinitely high, in case the untreated plants gave no yield, to zero, in case the treatment was phytotoxic and killed the plants. These extremes are rare but may occur. The result is usually a moderate yield increase for treated over untreated. The figure obtained in one particular trial is not suitable as a basis for generalization, due to great variability of trial results. The influence of soil (infestation, type, structure, fertility), plant (species, variety), treatment (chemical and dose, temperature and duration, ... etc.) and of environmental factors related to weather, moment of treatment, soil tillage before and after treatment and others, are great and unpredictable. This must have been the reason that hardly any estimate on the overall effect of PSS is available. It holds for the moment and may change as soon as the influential factors have been systematically analyzed and studied in detail.

In this chapter we approach the matter inductively by pooling and analyzing the results obtained with soil disinfection in The Netherlands during the period 1947-1970, i.e. from the moment field experiments with modern soil fumigants started in this country.

### 4.2. INVENTORY

All data within our reach on the effect of soil disinfection treatment on a certain plant in a certain soil in The Netherlands were collected, recalculated, tested, listed, inserted into computer cards and then used for making tables, diagrams and graphs to clarify and summarize the results. The total number of individual items was 2453.

The data were collected from different sources. They included published results on field and glasshouse experiments, about 1000 items sorted out from unpublished files of the PD, the LH, TNO-working parties and individual workers, and 545 items from experiments by the author. All data were included, apart from the exceptional extremes for which either the treated or the untreated yield was nil.

Pooling and statistical treatment of the heterogeneous data required standardization of the results. Each soil-plant-treatment combination was considered as an experimental item, for which the yield of the treated plants, or the average yield in case replicates were present, was expressed as a percentage of untreated. For most experiments several yield statistics were available; cf. 6.2.

TABLE 1. Some properties of the main soil sterilizers used.

Indication of common name or formulation used	Name and formula of active ingredient (= a.i.)	Physical form	Solubility in water, %	Boiling point in °C	Normal amount of a.i. in g per m <sup>2</sup>
CP = Chloropicrin	Trichloronitromethan = $\begin{array}{c} \text{Cl} \\   \\ \text{Cl}-\text{C}-\text{NO}_2 \\   \\ \text{Cl} \end{array}$	Yellowish liquid	0.23	112.4	50-80
MB = Methylbromide	Monobromomethan = $\begin{array}{c} \text{H} \\   \\ \text{H}-\text{C}-\text{Br} \\   \\ \text{H} \end{array}$	Colorless liquid	1.34	4.5	50-100
D = D D	1,3-dichloropropene = $\begin{array}{c} \text{H} & \text{H} & \text{H} \\   &   &   \\ \text{Cl}-\text{C}=\text{C}-\text{C}-\text{Cl} \\ & &   \\ & & \text{H} \end{array}$	Brownish liquid	0.1	95-150	17-40
EDB = Ethylenedi-bromide	1,2-dibromoethane = $\begin{array}{c} \text{H} & \text{H} \\   &   \\ \text{Br}-\text{C}-\text{C}-\text{Br} \\   &   \\ \text{H} & \text{H} \end{array}$	Colorless liquid	0.43	131.5	10-13
Trapex	Methylisothiocyanate = $\begin{array}{c} \text{H} \\   \\ \text{H}-\text{C}-\text{N}=\text{C}=\text{S} \\   \\ \text{H} \end{array}$	Light yellow liquid	0.67	118	18-22.5

Vapam	Sodium-N-methylthio- carbamate =	$  \begin{array}{c}  \text{H} \quad \text{N}-\text{H} \\    \quad   \\  \text{H}-\text{C}-\text{C}-\text{S}-\text{Na} \cdot 2\text{H}_2\text{O} \\    \quad   \\  \text{H} \quad \text{S}  \end{array}  $	72.2	-	30-40
Temik	2 methyl-2-(methylthio)-propion aldehyde-o-(methylcarbamoyl)-oxime =	$  \begin{array}{c}  \text{CH}_3 \\    \\  \text{N}-\text{C}-\text{O}-\text{N}=\text{CH}-\text{C}-\text{S}-\text{CH}_3 \\    \quad   \\  \text{H} \quad \text{CH}_3  \end{array}  $	0.6	-	0.15-0.9
CA = Captan	N-trichloromethylthio- 4-cyclohexene-1,2-dicarboximide =	$  \begin{array}{c}  \text{H}_2 \\    \\  \text{C} \\  / \quad \backslash \\  \text{HC} \quad \text{CH}-\text{CO} \\     \quad   \quad \backslash \\  \text{HC} \quad \text{CH}-\text{CO} \quad \text{NSC Cl}_3 \\  \backslash \quad / \\  \text{C} \\    \\  \text{H}_2  \end{array}  $	-	-	5-7.5
PCNB	Pentachloronitrobenzene =	$  \begin{array}{c}  \text{Cl} \\  / \quad \backslash \\  \text{Cl} \quad \text{Cl} \\    \quad    \quad   \\  \text{Cl} \quad \text{Cl} \quad \text{Cl} \\  \backslash \quad / \\  \text{NO}_2  \end{array}  $	-	-	4-14

In such cases the different statistics were usually correlated. The calculation was then based on the one statistic which best approximated the economical yield criterium used in agriculture.

Different formulations and doses of a chemical, or different temperatures and durations of a heat treatment, were initially kept separate. The basic list of 2453 experimental items comprises for each item: number, indication of the experiment including locality and year, soil type, crop, chemical and dose or other treatment specification, known soil-borne diseases or pests, and finally the plant yield on treated soil as a percentage of untreated. This voluminous list is not published here, but is kept available for consultation (EISSA 1971).

Before passing the cards through the computer a number of special programmes were made, which required further standardization or comprehension of the results into categories and also addition of special selection criteria. The following comprehensions or additions were applied for making the final tables and diagrams:

a. Of the more than 100 plant species on the basic list 28 important crops are recorded separately, and the remainder were grouped under the heading 'other crops'. This set-up was maintained in Tables 2-4 and in Fig. 6.

The crops mentioned specifically are: oat, wheat, barley, grass, clover, pea, bean, beet, potato, tomato, lettuce, endive, carrot, strawberry, rose, apple, malus, pear, prunus, black-currant, liguster, chrysanthemum, zinnia, narcissus, iris, gladiolus, hyacinth and other crops. Further specifications into species, variety and cultivar are recorded in the text if necessary.

b. Of the more than 30 disinfectants on the basic list (EISSA 1971) 10 important ones are recorded separately in Tables 2-4; the others are pooled under 'rest'. The disinfectants mentioned specifically are the nematocides dichloropropene (D), ethylene dibromide (EDB), the fungicides Captan and pentachloronitrobenzene (PCNB), the broadspectrum disinfectants heat (H), chloropicrin (CP) and methyl bromide (MB), and also Trapex and Vapam and Temik; cf. also Table 1. In Tables 2-4 no specifications were made for different doses, but this was elaborated on later as recorded under c.

c. The treatments with disinfectants or heat were always applied at one and often at several different doses. The influence of the doses is illustrated in Figs 2-4. They were indicated by the figures I, II, III, IV, V or VI, and the index was made so, that III approximated the normal amount recommended for practical application, II and IV were moderately low and moderately high respectively, I and V were low and high respectively. VI compiled all unidentified amounts and in some graphs the amount 0 (= untreated) was added. III represents the officially licensed or recommended amounts for application in The Netherlands, and the other categories were established after consulting with the Pesticides Department of the PD.

The doses I-V are recorded below as ml or g per m<sup>2</sup> of soil for the main sterilizers in Table 1, with the percentage of active ingredients indicated between brackets:



	I	II	III	IV	V
D (50%)	20-25 ml	30-40 ml	50-80 ml	100 ml	200 ml
EDB (41%)	15 ml	30 ml	45 ml	60 ml	120 ml
Heat		60°C	100°C	121°C	
CP (95%)		40-50 ml	80 ml	120 ml	
MB (98%)			75 g	100 g	
Trapex (24%)					150 ml
Vapam (50%)					100 ml
Temik (10%)			5 g		
Captan (83%)				100 g	
PCNB (20%)		16 g	40 g	80 g	

d. Many trials were laid on soil in which plant nematodes or other parasitic organisms were found before or during the experiment. The locating of a certain parasite does not prove that it has caused damage and that the effect of the soil treatment is due to its elimination. Also it does not prove that it is the only noxious organism in the soil. There is probably no soil without one or more parasitic organisms. However, the establishment of notable densities of pathogenic or suspected organisms enlarges the risk of damage and therefore the chance of obtaining positive soil disinfection effects. The following soil-borne pathogens or diseases were noticed, registered on the basic list of results (EISSA 1971) and summarized in Table 5:

*Heterodera* spp. (in general), *H. rostochiensis*, specific apple replant disease, *Pratylenchus penetrans*, the nematode complex *P. crenatus* + *Tylenchorhynchus dubius* + *Rotylenchus robustus*, *R. robustus*, *Hemicycliophora* + *Paratylenchus* spp., *Meloidogyne* spp., *Pythium* spp. There are additional categories for 'no pathogenic nematodes present' and for 'no infestation known'.

e. Eight soil types are specifically indicated in the study (BAKKER & SCHELLING 1966), namely loamy sand, clay, peaty sand, diluvial sand, dune sand, silt, polder sand and river sand, whereas other soils and soils of unknown composition have been grouped together under the heading 'other and unknown' (Table 8).

Detailed information on special cases will be given in the discussion if necessary. It should be kept in mind that the occurrence of certain plant parasites is closely correlated with soil type.

f. As most experiments were taken care of and registered by the district offices of the PD, the results were computed according to these districts namely Winschoten, Wageningen, Zwolle, Leiden, Utrecht, Rotterdam, Almelo, Exloo, Lisse and Emmeloord, with a remaining group indicated under 'Other locations' (Table 9).

g. The yield percentages of the treated compared to the untreated soils varied greatly. On the basis of practical considerations they were transformed into eight categories as follows:

-2 =  $\leq 70\%$ , -1 = 71-90%, 0 = 91-110%, 1 = 111-130%, 2 = 131-150%, 3 = 151-200%, 4 = 201-300%, 5 =  $> 300\%$  (cf. Figs 1 and 6).

The results of the inventory are summarized in Tables 2-9 and Figs 1-6.

Table 2 summarizes the overall results obtained with PSS, as well as the average result per treatment (11 treatments, including the rest group) and the average result per crop (29 crops, including the rest group), with reference to the number of experimental items on which the averages are based.

Table 3 and 4 are extracts from Table 2. Table 3 summarizes the same results as Table 2 if all items with figures below 100%, i.e. the treatments which caused yield depression, are omitted. Yield depression occurred in 481 of the 2453 items, and Table 3 thus comprises the results of 1972 items. The 481 toxic cases are calculated and presented according to the same scheme in Table 4.

The results obtained from each of the 11 treatments and each of the 29 crops are analyzed in Figs 1 and 6. The yield effects are divided into eight categories, and the frequency of each category is indicated. The diagram allows a comparison of the effect of PSS for each particular treatment and for each particular crop and thus demonstrates the success as well as the risk of PSS.

Figs 2-5 give further information on the risk of PSS, also related to doses. Fig. 2 analyzes the results obtained with the low, moderately low, normal, moderately high and high treatment doses. Fig. 3 illustrates the net practical result obtained for each dose by synthesizing the negative (yield-decreasing) and positive (yield-increasing) effects recorded in Fig. 2 into one index; it confirms that the choice of the normal dose, III, has been correct because it has caused a yield index as high as IV and superior to I, II and V. Fig. 4 relates the yield of the toxic cases to the corresponding non-toxic ones for each of the doses, and Fig. 5 demonstrates the relation between the occurrence of phytotoxic cases and the boiling point of the soil fumigant.

Tables 5-7, 8 and 9 relate the overall effect of PSS on the presence of soil-borne pests and diseases, to the soil type, and to different areas of the country, respectively.

### 4.3. DISCUSSION

#### 4.3.1. *General averages*

Tables 2-4 show, that PSS is a powerful, but also a risky technique to increase crop production, and that the differences between treatment-crop combinations are great. The yield on treated soil is 141% of untreated, as overall average for all treatments on all crops (2453 experiments).

If the toxic cases are excluded it is 154% (1972 experiments). The overall average of 141% has therefore been reached; 154% would have been reached if evident 'mistakes' by the experiments' executors had been avoided; the percentage which can potentially be reached if the best treatment and dose for each crop is chosen is much higher considering the many ineffective treatments occurring side by side with effective treatments in the same experiment. If the figures are calculated for all experiments with D, EDB, heat, CP, MB, Trapex and Vapam, which are the PSS treatments recognized in practice, 151%, or 164% without toxic cases, is obtained. If for each crop we select the kind of

TABLE 2. Computed average yield percentages of different crops on soils treated with different soil pesticides (untreated = 100). Between brackets are the numbers of experimental items on which the average percentages are based.

Crops	Soil treatments											Total
	D	EDB	H	CP	MB	Trapex	Vapam	Temik	Captan	PCNB	Rest	
Oat	125 (19)	- (-)	283 (2)	154 (11)	- (-)	- (-)	- (-)	162 (2)	102 (1)	99 (10)	- (-)	134 (45)
Wheat	117 (8)	- (-)	- (-)	- (-)	- (-)	131 (1)	125 (1)	- (-)	- (-)	- (-)	116 (5)	119 (15)
Barley	121 (8)	- (-)	172 (2)	196 (2)	- (-)	- (-)	- (-)	71 (2)	70 (2)	- (-)	- (-)	124 (16)
Grass	130 (42)	- (-)	135 (5)	133 (16)	- (-)	126 (1)	136 (1)	173 (1)	183 (1)	100 (15)	119 (5)	126 (87)
Clover	133 (28)	- (-)	- (-)	111 (11)	- (-)	- (-)	- (-)	- (-)	- (-)	93 (11)	143 (3)	121 (53)
Pea	165 (11)	- (-)	547 (2)	671 (1)	- (-)	- (-)	- (-)	386 (1)	429 (1)	- (-)	152 (3)	255 (19)
Bean	114 (19)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	114 (19)
Beet	108 (44)	- (-)	356 (4)	179 (13)	- (-)	115 (1)	110 (1)	- (-)	150 (1)	108 (11)	123 (6)	133 (81)
Potato	136 (61)	187 (7)	350 (1)	174 (14)	- (-)	111 (1)	114 (1)	382 (1)	232 (1)	103 (11)	135 (27)	144 (125)
Tomato	129 (38)	111 (13)	137 (4)	131 (6)	154 (5)	150 (3)	130 (13)	108 (1)	132 (1)	130 (1)	111 (76)	120 (161)
Cabbage	120 (18)	- (-)	229 (1)	227 (1)	- (-)	- (-)	- (-)	86 (1)	108 (1)	- (-)	- (-)	128 (22)
Lettuce	131 (14)	- (-)	- (-)	165 (11)	- (-)	- (-)	- (-)	- (-)	- (-)	121 (11)	- (-)	139 (36)
Endive	136 (9)	- (-)	101 (1)	142 (5)	- (-)	- (-)	144 (1)	- (-)	- (-)	107 (5)	109 (6)	125 (27)
Carrot	127 (65)	100 (1)	137 (5)	113 (18)	176 (2)	139 (8)	112 (19)	- (-)	- (-)	99 (15)	111 (57)	118 (190)
Rose	154 (60)	- (-)	237 (8)	129 (24)	119 (2)	196 (6)	162 (21)	- (-)	- (-)	88 (13)	193 (39)	153 (173)
Strawberry	117 (27)	- (-)	110 (8)	127 (13)	116 (1)	132 (4)	114 (7)	126 (2)	108 (4)	117 (8)	111 (22)	117 (96)
Apple	143 (81)	- (-)	460 (3)	223 (115)	183 (23)	148 (11)	135 (28)	- (-)	120 (21)	96 (10)	135 (98)	166 (390)
Malus	151 (48)	- (-)	- (-)	139 (1)	127 (4)	160 (7)	156 (16)	- (-)	- (-)	99 (1)	129 (41)	152 (118)
Pear	140 (8)	- (-)	- (-)	165 (7)	149 (2)	- (-)	140 (2)	- (-)	- (-)	- (-)	125 (8)	143 (27)
Prunus	207 (25)	- (-)	- (-)	- (-)	- (-)	172 (4)	180 (8)	- (-)	- (-)	- (-)	161 (20)	185 (57)
Black-currant	104 (12)	- (-)	- (-)	- (-)	- (-)	105 (2)	118 (5)	- (-)	- (-)	- (-)	115 (12)	110 (31)
Liguster	97 (17)	- (-)	- (-)	96 (8)	- (-)	- (-)	163 (2)	- (-)	- (-)	111 (8)	126 (12)	110 (47)
Chrysanthemum	116 (11)	- (-)	- (-)	123 (11)	- (-)	- (-)	- (-)	- (-)	- (-)	106 (10)	- (-)	115 (32)
Zinnia	103 (7)	- (-)	- (-)	105 (7)	- (-)	- (-)	- (-)	- (-)	- (-)	99 (7)	- (-)	103 (21)
Narcissus	122 (8)	120 (5)	- (-)	- (-)	- (-)	118 (3)	129 (8)	- (-)	- (-)	- (-)	121 (11)	123 (35)
Iris	102 (6)	- (-)	- (-)	- (-)	- (-)	110 (2)	148 (13)	- (-)	- (-)	- (-)	111 (9)	125 (30)
Gladiolus	118 (5)	- (-)	- (-)	- (-)	- (-)	110 (1)	107 (1)	- (-)	- (-)	- (-)	99 (5)	109 (12)
Hyacinth	- (-)	- (-)	- (-)	- (-)	- (-)	110 (1)	116 (55)	- (-)	- (-)	- (-)	109 (140)	111 (196)
Other crops	187 (207)	- (-)	- (-)	153 (3)	147 (2)	124 (18)	125 (15)	- (-)	- (-)	72 (1)	148 (46)	168 (292)
Total	146 (906)	133 (26)	221 (46)	173 (298)	166 (41)	141 (74)	132 (218)	168 (11)	130 (34)	103 (148)	124 (651)	141 (2453)

TABLE 3. Computed average yield percentages for the data of Table 2 exclusive toxic cases with yields lower than untreated.

Soil treatments		D	EDB	H	CP	MB	Trapex	Vapam	Temik	Captan	PCNB	Rest	Total
Crops													
Oat	130 (16)	- (-)	283 (2)	162 (10)	- (-)	- (-)	- (-)	- (-)	162 (2)	102 (1)	104 (5)	- (-)	145 (36)
Wheat	125 (6)	- (-)	- (-)	- (-)	- (-)	- (-)	131 (1)	126 (1)	- (-)	- (-)	- (-)	121 (4)	124 (12)
Barley	126 (7)	- (-)	172 (2)	196 (2)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	147 (11)
Grass	134 (39)	- (-)	135 (5)	133 (16)	- (-)	- (-)	126 (1)	136 (1)	173 (1)	183 (3)	105 (9)	125 (4)	131 (77)
Clover	147 (21)	- (-)	- (-)	115 (9)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	103 (3)	143 (3)	135 (36)
Pea	256 (6)	- (-)	547 (2)	671 (1)	- (-)	- (-)	- (-)	- (-)	386 (1)	429 (1)	- (-)	152 (3)	326 (14)
Bean	136 (12)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	136 (12)
Beet	133 (24)	- (-)	441 (3)	216 (9)	- (-)	115 (1)	111 (1)	110 (1)	- (-)	150 (1)	131 (5)	128 (5)	166 (49)
Potato	143 (53)	205 (6)	350 (1)	180 (13)	- (-)	111 (1)	114 (1)	114 (1)	382 (1)	232 (1)	113 (6)	156 (21)	156 (104)
Tomato	137 (34)	113 (11)	137 (4)	131 (6)	154 (5)	150 (3)	133 (12)	133 (12)	108 (1)	132 (1)	130 (1)	116 (59)	126 (137)
Cabbage	134 (13)	- (-)	229 (1)	227 (1)	- (-)	- (-)	- (-)	- (-)	- (-)	108 (1)	- (-)	- (-)	144 (16)
Lettuce	141 (11)	- (-)	- (-)	165 (11)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	184 (4)	- (-)	158 (26)
Endive	143 (8)	- (-)	101 (1)	142 (5)	- (-)	- (-)	- (-)	144 (1)	- (-)	- (-)	114 (4)	110 (5)	130 (24)
Carrot	139 (50)	100 (1)	166 (3)	127 (12)	176 (2)	158 (6)	121 (14)	121 (14)	- (-)	- (-)	108 (7)	118 (46)	129 (141)
Rose	164 (53)	- (-)	258 (7)	140 (19)	119 (2)	215 (5)	177 (17)	177 (17)	- (-)	- (-)	121 (5)	171 (33)	168 (141)
Strawberry	126 (20)	- (-)	115 (6)	129 (12)	116 (1)	132 (4)	122 (6)	122 (6)	126 (2)	113 (3)	148 (4)	120 (17)	125 (75)
Apple	152 (70)	- (-)	460 (3)	232 (108)	183 (23)	170 (8)	144 (24)	144 (24)	- (-)	129 (17)	104 (5)	162 (74)	180 (332)
Malus	162 (41)	- (-)	- (-)	139 (1)	142 (3)	160 (7)	161 (15)	161 (15)	- (-)	- (-)	- (-)	163 (35)	162 (102)
Pear	140 (8)	- (-)	- (-)	165 (7)	149 (2)	- (-)	- (-)	140 (2)	- (-)	- (-)	- (-)	136 (6)	147 (25)
Prunus	229 (21)	- (-)	- (-)	- (-)	- (-)	- (-)	172 (4)	180 (8)	- (-)	- (-)	- (-)	165 (19)	194 (52)
Black-currant	134 (6)	- (-)	- (-)	- (-)	- (-)	116 (1)	136 (3)	136 (3)	- (-)	- (-)	- (-)	133 (7)	133 (17)
Liguster	113 (8)	- (-)	- (-)	114 (4)	- (-)	- (-)	163 (2)	163 (2)	- (-)	- (-)	122 (5)	134 (10)	125 (29)
Chrysanthemum	118 (10)	- (-)	- (-)	123 (11)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	113 (6)	- (-)	119 (27)
Zinnia	109 (5)	- (-)	- (-)	113 (4)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	106 (4)	- (-)	109 (13)
Narcissus	122 (8)	126 (4)	- (-)	- (-)	- (-)	118 (3)	129 (8)	129 (8)	- (-)	- (-)	- (-)	121 (11)	123 (34)
Iris	110 (4)	- (-)	- (-)	- (-)	- (-)	110 (2)	148 (13)	148 (13)	- (-)	- (-)	- (-)	119 (7)	132 (26)
Gladiolus	118 (5)	- (-)	- (-)	- (-)	- (-)	110 (1)	107 (1)	107 (1)	- (-)	- (-)	- (-)	115 (2)	115 (9)
Hyacinth	- (-)	- (-)	- (-)	- (-)	- (-)	110 (1)	118 (50)	118 (50)	- (-)	- (-)	- (-)	114 (112)	115 (163)
Other crops	218 (162)	- (-)	- (-)	192 (2)	147 (2)	128 (16)	125 (15)	125 (15)	- (-)	- (-)	- (-)	125 (35)	191 (232)
Total	162 (721)	140 (22)	240 (40)	184 (263)	168 (40)	148 (65)	138 (195)	138 (195)	203 (8)	143 (27)	118 (73)	133 (518)	154 (1972)

TABLE 4. Computed average yield percentages for toxic cases in Table 2

Crops	Soil treatments										Rest	Total
	D	EDB	H	CP	MB	Trapez	Vapam	Temik	Captan	PCNB		
Oat	95 (3)	- (-)	- (-)	72 (1)	- (-)	- (-)	- (-)	- (-)	- (-)	94 (5)	- (-)	92 (9)
Wheat	96 (2)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	99 (1)	97 (3)
Barley	85 (1)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	71 (2)	70 (2)	- (-)	- (-)	73 (5)
Grass	81 (3)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	93 (6)	94 (1)	89 (10)
Clover	90 (7)	- (-)	- (-)	93 (2)	- (-)	- (-)	- (-)	- (-)	- (-)	89 (8)	- (-)	90 (17)
Pea	56 (5)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	56 (5)
Bean	75 (7)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	75 (7)
Beet	78 (20)	- (-)	98 (1)	95 (4)	- (-)	- (-)	- (-)	- (-)	- (-)	89 (6)	99 (1)	84 (32)
Potato	90 (8)	83 (1)	- (-)	98 (1)	- (-)	- (-)	- (-)	- (-)	- (-)	92 (5)	78 (6)	87 (21)
Tomato	68 (4)	99 (2)	- (-)	- (-)	- (-)	- (-)	92 (1)	- (-)	- (-)	- (-)	94 (17)	90 (24)
Cabbage	83 (5)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	86 (1)	- (-)	- (-)	- (-)	84 (6)
Lettuce	97 (3)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	86 (7)	- (-)	89 (10)
Endive	77 (1)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	79 (1)	98 (1)	85 (3)
Carrot	85 (15)	- (-)	93 (2)	86 (6)	- (-)	84 (2)	87 (5)	- (-)	- (-)	91 (8)	80 (11)	85 (49)
Rose	85 (7)	- (-)	92 (1)	89 (5)	- (-)	99 (1)	97 (4)	- (-)	- (-)	67 (8)	85 (6)	83 (32)
Strawberry	90 (7)	- (-)	97 (2)	94 (1)	- (-)	- (-)	67 (1)	- (-)	92 (1)	86 (4)	81 (5)	87 (21)
Apple	85 (11)	- (-)	- (-)	86 (7)	- (-)	88 (3)	78 (4)	- (-)	81 (4)	87 (5)	85 (58)	85 (58)
Malus	86 (7)	- (-)	- (-)	- (-)	82 (1)	- (-)	89 (1)	- (-)	- (-)	99 (1)	89 (6)	88 (16)
Pear	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	93 (2)	93 (2)
Prunus	91 (4)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	82 (1)	89 (5)
Black-currant	74 (6)	- (-)	- (-)	- (-)	- (-)	93 (1)	90 (2)	- (-)	- (-)	- (-)	91 (5)	84 (14)
Liguster	83 (9)	- (-)	- (-)	78 (4)	- (-)	- (-)	- (-)	- (-)	- (-)	93 (3)	92 (2)	84 (18)
Chrysanthemum	89 (1)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	94 (4)	- (-)	93 (5)
Zinnia	89 (2)	- (-)	- (-)	94 (3)	- (-)	- (-)	- (-)	- (-)	- (-)	91 (3)	- (-)	92 (8)
Narcissus	- (-)	96 (1)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	97 (1)
Iris	87 (2)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	83 (2)	85 (4)
Gladiolus	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	89 (3)	89 (3)
Hyacinth	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	97 (5)	- (-)	- (-)	- (-)	94 (28)	95 (33)
Other crops	77 (45)	- (-)	- (-)	77 (1)	- (-)	97 (2)	- (-)	- (-)	- (-)	72 (1)	87 (11)	79 (60)
Total	82 (185)	94 (4)	95 (6)	88 (35)	82 (1)	91 (9)	89 (23)	76 (3)	79 (7)	87 (75)	88 (133)	86 (481)

PSS treatment which was on average the most effective and combine the results, an overall average of 172%, based on 421 experiments or 188% without toxic cases, based on 385 experiments is reached. These figures would become even higher if we took into account that the best treatments for each crop would still be an average, and that other treatments would have been more efficient on particular soils or in particular trials. It is therefore safe to conclude, that PSS has demonstrated the power to increase the yield of crops in The Netherlands to more than 172%, or to 188% if toxic cases are excluded, on soils similar to those used for our experiments. This effect is of the same magnitude as the effect of 187% recorded by TAYLOR (1961) for the U.S.A.

To further evaluate the results some points must be stressed. Firstly, it should be kept in mind that the data in Tables 2-4 are collected from experiments. About half the fields were selected either because they contained familiar problems or because they harboured organisms, known or suspected to be plant parasitic; and for this reason the results of PSS observed must be higher than would have been obtained on randomly chosen soils. This problem of selectivity will be further dealt with in the following chapters. Secondly: possible fertilizing effects of the treatments, on the other hand, were compensated to a larger extent than would have occurred under practical conditions, because the large number of PSS-trials conducted by workers at the PD including the author usually received about twice the normal amount of fertilizers. Finally: among the experimental results there were many cases of toxic effects, namely 481 out of 2453, which is about 20%. The separation of toxic from non-toxic cases at 100%, i.e. the level of untreated, disregards the influence of experimental variability, but is acceptable in this statistical approach. It is, however, important to know whether the percentage of toxic cases observed in all these experiments could be expected in practice if PSS was applied. Application of PSS will be done with less precision and knowledge by practical farmers than in the case of our experiments. On the other hand it is certain that several of the experiments on our list were carried out with new or unusual chemicals or with extreme doses and that such treatments will not be recommended for practical use. Phytotoxicity will probably be less common in practice than in our experiments. The matter of phytotoxic effects will further be dealt with under 4.3.2.

The results obtained, therefore, indicate that PSS increases production of all kinds of crops on many soils. The yield increase on problem fields is drastic. Without doubt the figures are above average for normal soil, but may be below average for soils under practical treatment because a large number of randomly chosen trial fields are included in our survey. It is probable that the number of toxic cases will in practice be lower than was found here, and therefore that the figures of Table 3 are more representative for the results on problem fields than those in Table 2. In the following text we shall frequently be mentioning side by side averages inclusive of the toxic cases and averages without them.

### 4.3.2. Treatment averages

Sometimes the treatment averages in Tables 2-4 and the analyzed results in Fig. 1 had to be completed with data from the unpublished basic list of individual trial results, especially so when few experiments per treatment had been available and the risk, that one or a few soils had biased the results, would be great.

The average yields obtained were, in increasing order, 103% for PCNB, 124% for the complex remaining group of treatments, 130% for Captan, 132% for Vapam, 133% for EDB, 141% for Trapex, 146% for D, 166% for MB, 168% for Temik, 173% for CP and 221% for heat. When the toxic cases are excluded, the figures are 118% for PCNB, 133% for the rest group, 138% for Vapam, 140% for EDB, 143% for Captan, 148% for Trapex, 162% for D, 168% for MB, 184% for CP, 203% for Temik, and 204% for heat.

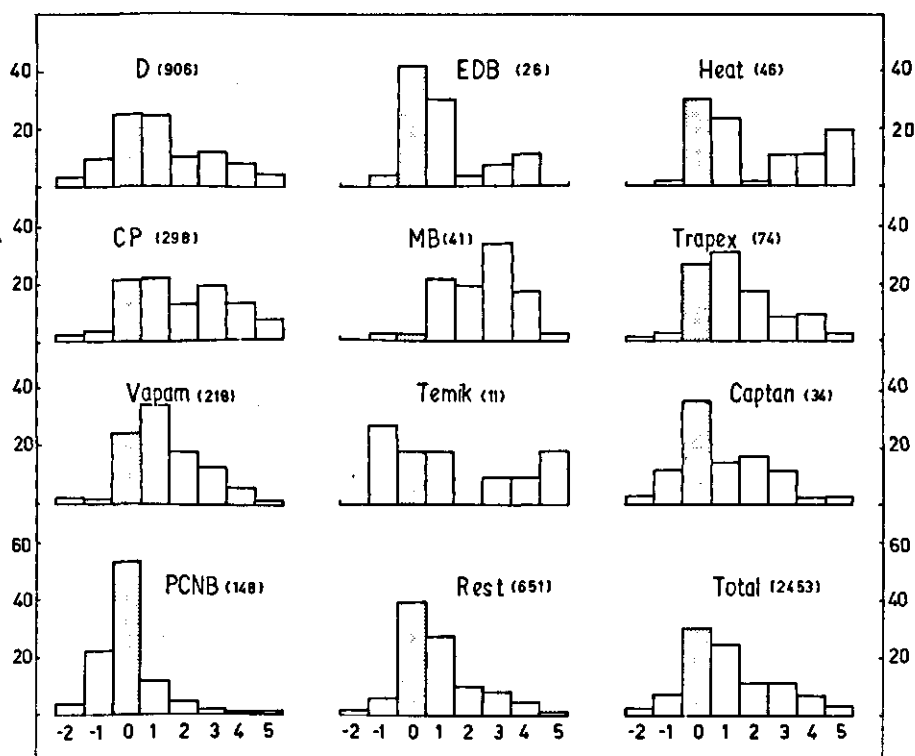


FIG. 1. Frequency of different yield results obtained by PSS with different treatments irrespective of dose applied or crop. The results are presented as eight different categories: -2 =  $\leq 70\%$  (i.e. yield of treated is  $\leq 70\%$  of untreated), -1 = 71-90%, 0 = 91-110%, 1 = 111-130%, 2 = 131-150%, 3 = 151-200%, 4 = 201-300%, 5 =  $> 300\%$ . Abscissa for each treatment diagram: categories indicating average yield results obtained by PSS. Ordinate for each treatment diagram: frequencies indicating percentage of total number of experiments for the treatment; this number is added between brackets.

The results for EDB, MB, Temik and Captan will first be discussed because they are all based on only a few experiments, and because the figures from the basic list confirm that the results are, or may be, special and not general averages.

### *EDB*

The results for EDB seem to be less than for D, although both are known to be good nematicides, widely used all over the world. There were only 26 experimental items for EDB. The 7 experiments for potato all concern control of *Heterodera rostochiensis* in heavily infested dune sand reaching the high score of 187%. The 5 treatments of narcissus were about as effective as for D.

There were a further 13 experiments of tomato, in fields where the effect of PSS was generally low, and one treatment of carrot without effect. Examination of the results, therefore, does not substantiate the impression that EDB is less effective than D. The analysis in Fig. 1 confirms that EDB may increase the yield to about the same degree as D.

### *MB*

MB scores a higher average than D, namely 166%/168% against 146%/162%. The MB averages, however, are based on only 41 experiments. 23 of them concern the partly unidentified apple replant disease in which a high score of 183%/183% was reached; this explains the top at the yield increase index 3 in Fig. 1. When this one disease is left out of scope, MB appears to have been less effective than D.

### *Temik*

Temik was applied in only 11 experiments, but here it is recorded separately because it reached the very high score of 168%/203%, and because it is the only soil pesticide on our list which is systemic and can be applied directly to growing crops. Little is known about the effects of this new chemical, but the results indicate that it may be a broad-spectrum pesticide with effects comparable to those of heat or CP. Or that it has unidentified specific growth-promoting properties. The diagram in Fig. 1 reflects the ability for increasing yields, quite heavily in some cases, as well as the fact that phytotoxic effects are neither absent, nor exceptional if the chemical is directly applied to crops; cf. also Chapter 6.

### *Captan*

Captan is known as a fungicide, but later experiments have also shown that it may act as a nematicide at the doses used (cf. Chapter 6). The list summarized in Tables 2-4 comprises 34 experiments and the variations between the effects are great. This can also be read from the diagram in Fig. 1. Captan reached an average yield of 130%/143%, mainly owing to some very high effects in incidental experiments on pea and potato and a fairly high effect in 21 experiments against apple replant disease.



The sterilizers for which the number of experiments is sufficient to consider the average to be of general significance, are: D which is known as a nematicide, heat and CP which are broad-spectrum treatments, Trapex and Vapam which are often indicated as broad-spectrum disinfectants but do not show such power in these results, PCNB which is considered a fungicide, and the remaining heterogeneous group.

## D

The different formulations of dichloropropene reach an average yield of 146%/162%; 185 of the 906 experiments are toxic cases. This is not above the overall average, but it is nevertheless high because D is the best-known and most widely used chemical for PSS, and the many toxic cases stress the fact that PSS is still a risky technique. As indicated before, most of the toxic cases would probably have been avoided in practical applications. The cost of a D treatment may be estimated at f 400–500/ha. A general treatment with D of all soils used in our experiments would have given a much higher financial return. This result suggests that the application of D in Dutch agriculture is probably economic on many soils. The analysis in Fig. 1 indicates that about 2/3 of all experiments gave a favourable response, and that more than 1/5 showed yield increases of more than 50%.

## Heat

Heat caused the highest yield increase of all treatments, probably because it killed all noxious organisms and released more fertilizer components than other treatments. It reached 221%/240%. It was usually, but not always superior to D, and it may solve problems which D can not or only partially solve. The extra effect of heat above D in oat and barley, pea, beet, potato, rose and apple suggests that these crops suffer from soil-borne fungi and that this damage occurs independently from nematode damage. The analysis in Fig. 1 confirms, that heat may solve serious problems not solved by D, and also that heat does not cause many toxic cases, although they are present. The cost of a heat treatment is about 10 times the cost of a D treatment, and therefore prohibits application in practice except for small amounts of soil in which precious crops have to be grown.

## CP

The effect of CP is approximately comparable to that of heat; it reached 173%/184%. This is somewhat less than for heat, but CP was incorporated in a large series of field trials on soils without marked yield deficiencies, whereas heat was not. This explains the difference between the average results. CP also caused few toxic cases, as was the case for heat. The analysis in Fig. 1 shows, that strong responses occurred in a high percentage of the experiments. CP, therefore, can solve problems as well as heat, and is superior to D in many soils. The cost, which is 5–10 times that of D, restricts practical application to special cases.

### *Trapex and Vapam*

Trapex and Vapam reach 141%/148% and 132%/138% respectively. Their effects are similar for nearly all crops, which could be expected because the active ingredient of Trapex after transformation in the soil is methylisothiocyanate, the same as for Vapam. Both chemicals are often recommended as broad-spectrum soil pesticides, but this is not substantiated from our results. Their effect is generally comparable to that of the nematicides D and EDB, and not to heat or CP which are effective against the host specific problems in pea, apple and some other plants. This does not exclude the possibility that methylisothiocyanate can control some particular problems caused by fungi or bacteria. Trapex and Vapam in general, however, appear to be zoocides, comparable to D and EDB, rather than fungicides. The diagrams in Fig. 1 confirm that Trapex and Vapam often cause moderate yield increases and seldom reach the strong effects of heat and CP.

### *PCNB*

PCNB is recognized as a fungicide. It reached only 103%/118%. These figures are low and they also indicate that PCNB was often toxic: 75 out of 148 experiments were toxic cases. It is noteworthy that the average percentage effect of PCNB approximated the differences between the effects reached by CP and D. Table 3, recording the effects without toxic cases, reveals that PCNB had a notable positive effect in several experiments on beet, lettuce, and strawberry. The analysis of the results in Fig. 1 confirms that PCNB treatments are usually ineffective and often toxic, and therefore that it is not promising for PSS in general with the doses used.

### *Rest*

The rest of the treatments, comprising a diverse group of chemicals, reached an average yield of 124%/133%. The distribution of the effects is much the same as for D, Trapex and Vapam. There are promising treatments in this group, but they are not discussed separately because the number of experiments was too small for general evaluation.

### *Total*

The effects of all treatments, together reached 141%/154%, with about 20% toxic cases. The analysis of Fig. 1 reveals a distribution of the effects which closely resembles that of D. The frequencies reached by the different categories were 61 for -2 (<70% yield), 186 for -1 (71-90%), 748 for 0 (91-110%), 626 for 1 (111-130%), 285 for 2 (131-150%), 284 for 3 (151-200%), 177 for 4 (201-300%) and 86 for 5 (>300%).

Comparison of the detailed effects of the treatments used, therefore, leads to the following conclusions: the fungicide PCNB was ineffective and risky. The fungicide Captan (which has some nematicidal properties) was sometimes effective but also risky. The well known nematicides D, Trapex and Vapam,

increased the yields on many soils to a considerable extent, with more toxic cases for D than for the others. The broad-spectrum soil disinfectants CP and heat reached higher yield percentages than the other categories and did not cause many toxic cases. The results for EDB, MB, and Temik were not based on many experiments, but scrutiny of all results including those on the basic list, indicates that EDB is usually not less and MB not better than D (as Fig. 1 and the index mentioned later would suggest), and that Temik may appear to be a broad-spectrum sterilant with the same ability as CP or heat. The indication that MB, Trapex and Vapam are in practice rather zoocides comparable to D and EDB than broad-spectrum sterilants compared to CP and heat, is unexpected considering the literature review in Chapter 2. Assuming that the experimental results represent practical applications, we could illustrate the net practical result obtained with each treatment by synthesizing the negative (yield-decreasing) and positive (yield-increasing) effects according to the diagrams of Fig. 1 into one index. Such an index could be the product of 'yield categories'  $\times$  'corresponding frequency percentages'. These indices for the treatment-diagrams of Fig. 1 are, in the order of increasing values, PCNB = 2, 'Rest' = 88, CA = 94, EDB = 104, 'Total' = 117, D = 122, Vapam = 129, Trapex = 137, T = 146, CP = 186, H = 200 and MB = 241. This 'net practical result index' has only a comparative value and it is subjective in this respect that toxic cases cause a rapid fall and high yield increases a steep rise of the index. It shows nevertheless a logical sequence if we accept as bases the yield results of our experiments. Scrutiny of the incidental data indicates that the index is low for EDB and high for MB due to some exceptional results, as indicated before.

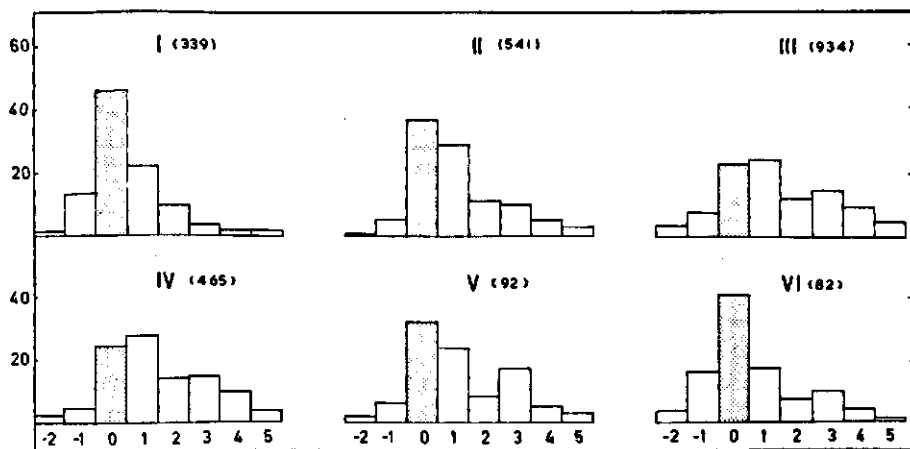


FIG. 2. Frequency of different yield results obtained by PSS with the low (I), moderately low (II), normal (III), moderately high (IV), high (V), and unidentified treatment doses (VI), based on all yield results irrespective to kind of treatment or crop. Abscissa for each dose diagram: categories indicating average yield results obtained by PSS. Ordinate for each dose diagram: frequencies indicating percentage of total number of experiments for the dose; this number is added between brackets.

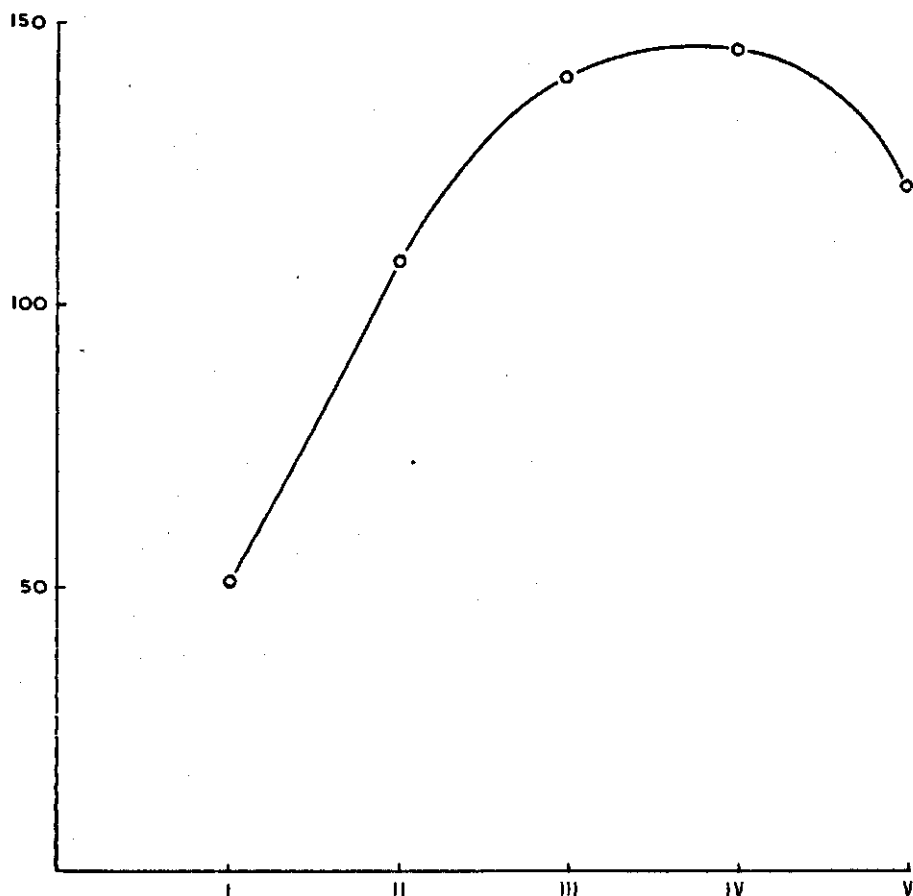


FIG. 3. Values calculated from the data of Fig. 2. For each PSS dose the product of 'yield categories'  $\times$  'corresponding frequency percentages' is calculated, thus yielding index for the net practical result obtained with each dose. Abscissa: doses I-V as indicated in Fig. 2. Ordinate: yield index calculated.

The effect of a treatment usually depends on the dose, indicated by category numbers, I-V. Fig. 2 summarizes the frequency of different yield results by PSS at low (I), moderately low (II), normal (III), moderately high (IV), and high (V) doses, complemented by a category for unidentified doses (VI) which will further not be discussed. Fig. 3 illustrates the 'net result index', calculated in the same way as was done earlier for the different treatments, by determining the product of the 'yield categories'  $\times$  'corresponding frequency percentages'. Figs 4 and 5 further elaborate on phytotoxicity.

At first sight, there seems little difference between the effects from the doses, but scrutiny of the diagrams in Fig. 2 reveals essential differences. The percentages of toxic cases is higher for I, the lowest dose, than for II-V, possibly because for the lowest dose phytotoxicity was often not compensated by

FIG. 4. Relation between the applied PSS doses and the yield of toxic cases as percentages of corresponding non-toxic cases. Abscissa: applied PSS doses I-V as in Fig. 2. Ordinate: yield of toxic cases as percentage of corresponding non-toxic cases.

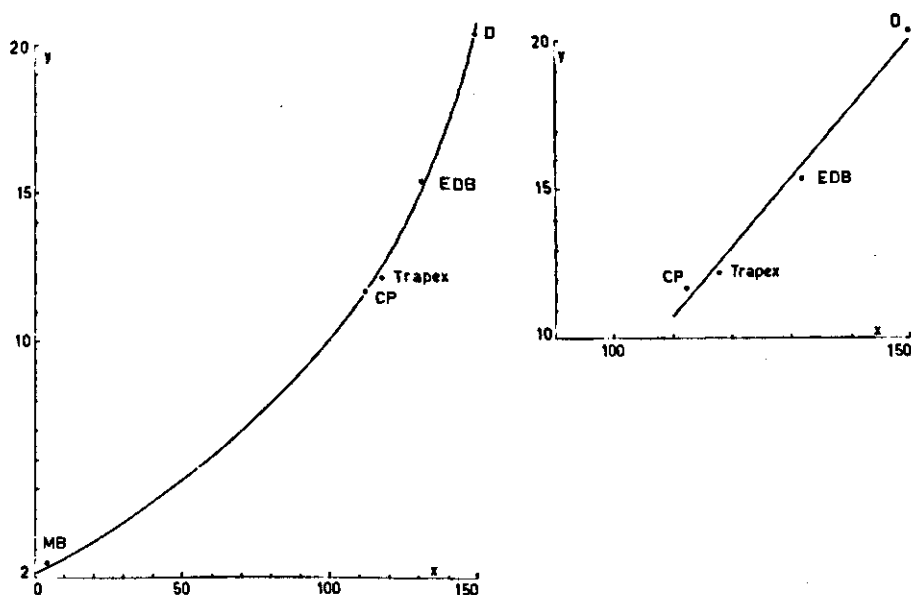
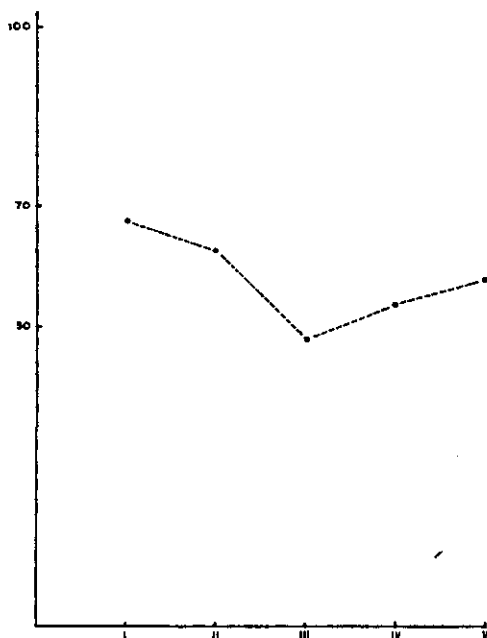


FIG. 5. At left: Relation between boiling point of the main fumigants used and the percentage toxic cases caused by them. At right: Linear regression for four fumigants with boiling points between 100-150°C. Formula:  $y = 0.237 x - 15.43$ , regression coefficient  $r = 0.993^{**}$ . Abscissas: X = boiling point in °C. Ordinates: Y = percentage toxic cases of total number of experiments.

sufficient PSS effect. Yield increases, especially high ones, occurred more frequently when the doses increased from I to III, with perhaps some further increase at IV, but with less good results at V. This is substantiated further in Fig. 3. It appears that the normal doses (III) caused a yield index as high as IV and was superior to I, II and V. The choice of III as the normal, i.e. the practically recommended dose, appears to be correct. IV reaches the same result, but it is less economic. V is even inferior, which must be due to phytotoxicity.

It is likely from these results that successful treatments with a yield figure above 100% often suffer from phytotoxicity which is then overcompensated by the positive, yield increasing effect. The results of toxic cases may as well be a result of growth promotion and toxic effect, with in these cases toxic effect dominating. Fig. 4 indicates that the percentage yield of toxic cases compared to non-toxic cases is not or not regularly influenced by the dose which also suggests that phytotoxicity and growth promotion are, simultaneously, influenced by dose.

Phytotoxicity is one of the major limiting factors in the application of PSS. It is due to slow disappearance of the chemicals, which are mostly evaporating liquids. Fig. 5 reveals the highly significant relation between number of cases with toxic effects in our experiments and boiling point of the main fumigants used. The percentage toxic cases increases markedly, from negligible to above 20%, for MB, CP, Trapex, EDB, D, in that order and the boiling point increases correspondingly from 4 to 150°C. For the chemicals CP, Trapex, EDB and D, all with boiling points between 112 and 150°C, a linear regression line is drawn and its formula is indicated in Fig. 5. This great difference in phytotoxic effect must have markedly suppressed the yield figures for D in comparison to MB. This holds for the results of our survey, but may also be true in practical applications.

#### 4.3.3. Crop averages

The crop averages and other results in Tables 2-4 and Fig. 6 will sometimes have to be completed with information from the basic list of individual trial results. This is certainly the case when few experiments per crop are available, because the choice of one or a few soils may then have influenced the final results too much and therefore have biased the average. Each crop average, or incidentally, group average, will therefore have to be considered critically.

The extremely high average for pea and the extremely low figures for zinnia, black-currant and liguster are not representative averages, although they reveal interesting phenomena of PSS. They will be discussed first.

#### Pea

This crop reached 255%/326% (19/14 experiments). The high result, however, is mainly due to one trial with different treatments on a clay soil infested with *Heterodera goettingiana* where peas had been cultivated for several years in succession, so that also other pathogenic nematodes and fungi had probably accumulated. The treatments in this trial (cf. Table 14 for details) were D,

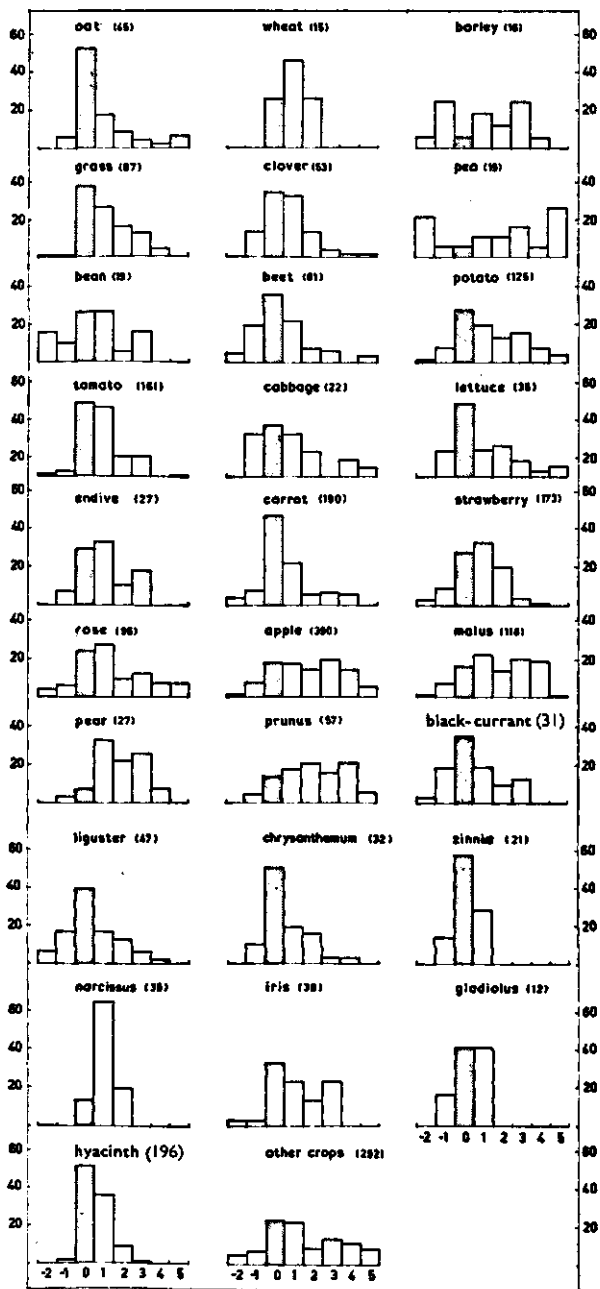


FIG. 6. Frequency of different yield results obtained with PSS on individual crops irrespective of kind of treatment. The results are presented as eight different categories:  $-2 = \leq 70\%$  (i.e. yield of treated is  $\leq 70\%$  of untreated),  $-1 = 71-90\%$ ,  $0 = 91-110\%$ ,  $1 = 111-130\%$ ,  $2 = 131-150\%$ ,  $3 = 151-200\%$ ,  $4 = 201-300\%$  and  $5 = > 300\%$ . Abscissa for each crop diagram: categories indicating average yield results obtained by PSS. Ordinate for each crop diagram: frequencies indicating percentage of total number of experiments for the crop; this number is added between brackets.

heat, CP, Temik and Captan, and the results were 816%, 886%, 671%, 386% and 429%, which excessively, influenced the average of the only 19 experiments. It is remarkable that pea also reached the maximum figure for phytotoxicity, namely 56% yield for toxic cases, which was the lowest average in Table 4. This is due to the DD-treatments only: 5 of the 11 DD experiments were phytotoxic to such an extent, that pea must be considered extremely susceptible to DD remnants in the soil (cf. also Fig. 6).

### *Zinnia*

There were only 21 treatments for zinnia and the result was only 103%/109% on average. The reason is probably that zinnia was grown only as a test plant on soils where the plant had never been grown before. No treatment in any of the 12 test soils exceeded +1 or -1 in Fig. 6.

### *Black-currant*

There were only 31 treatments for this plant and the result was 110%/133%. Phytotoxicity obviously influenced the results strongly. The second figure, effect without toxic cases and the diagram in Fig. 6, is therefore useful as an estimate. Black-currant was grown in one field where 9 treatments resulted in 9 toxic cases with an average yield percentage of 91 and in another field where the same 9 treatments caused 3 toxic cases with a total average of 103; in other fields several treatments resulted in incidental percentages varying between 110 and 177. Black-currant is grown as unrooted cuttings and these are now known to be very susceptible to several soil disinfectants. The time between treatment and planting has been too short in the two first mentioned trials with many treatments, and the resulting damage has biased the general average.

### *Liguster*

Liguster reached 110%/125% and the general average is also strongly suppressed due to 18 phytotoxic cases out of a total of 47. Liguster is also planted as unrooted cuttings and is very susceptible to phytotoxic residues of soil sterilants, as was black-currant, cf. also Fig. 6.

The other crops will be discussed in the order of Tables 2-4.

### *Oat*

PSS increased the yield to 134%/145%, which is considerable. 9 out of 45 experimental items were phytotoxic cases, which conforms to the average. The result by D, which was often applied, was 125%/130%, but in a few experiments much higher figures were obtained with heat, CP and Temik. This suggests, that soil fungi caused damage to oat, in addition to or in association with nematodes. The specific fungicides Captan and PCNB, on the other hand, did not have noticeable results according to this inventory. The diagram in Fig. 6 indicates, that PSS often has little effect on oat, but that the effect is extremely great in several other cases. Oat, therefore, suffers markedly in a number of soils. The effect of PSS on oat is studied in more detail later (cf. 6.9. and 8).



### *Wheat*

Wheat reached 119%/124%, which is a lower result than for the other cereals. There were 15 experimental items and this number is too low for a general estimate. This is especially so because the treatments were made on soils where wheat was not normally grown. Fig. 6 suggests also that no soils with serious problems for wheat were used, for the yield increase did not exceed the index 2. There was no measurable phytotoxicity.

### *Barley*

For barley 124%/147% was reached. Comparison of the results in Fig. 6 with those for oat suggests that barley suffers more often and more seriously than oat, but also that toxic cases occur more often. The few treatments with heat and CP were superior to D. This suggests that for barley fungi cause damage in addition to nematodes as well.

### *Grass*

PSS caused yield increases for grass, mainly English ryegrass, to 126%/131%. The difference between D and broad-spectrum disinfectants was negligible. There were very few toxic cases; apparently grass is not very susceptible to soil pesticides in general. Fig. 6 indicates, that PSS hardly influences grass growth on about 40% of the soils, but that considerable yield deficiencies occur on many other soils. The result is low compared to previously published results. OOSTENBRINK (1954) reported an average yield increase of 77% for meadows. It must, however, be kept in mind that our experiments only as exceptional cases were carried out on soils on which grass had been grown before. The effect of PSS on the growth of grass will be examined in greater detail later (cf. 6.4. and 7.4.).

### *Clover*

The clover studied was mostly white clover, but the results on red clover are added because the crops behaved similarly. PSS increased the yield to 121%/135%. 17 out of 53 experimental items were toxic cases, which is a high percentage. D was superior to CP and other treatments, which suggests that no fungus infestations are involved. Fig. 6 indicates, that PSS did not influence clover on about 35% of the soils, whereas extremely high yield deficiencies occurred on a number of soils. It should be noted, as for grass, that most of the soils used had not been under clover before.

### *Bean*

*Phaseolus* and *Vicia* were comprised under this heading. PSS, in all cases with D, increased the yields to 114%/136%. The percentage of toxic cases, due to damage by D, was high as for other Leguminosae.

### *Beet*

Beet reached 133%/166%. This is a high score, but it is clear from these

figures, and also from Fig. 6, that many phytotoxic cases occurred, especially due to D. It appears from the figures of Table 3, that heat and CP, and also the fungicides Captan and PCNB, were superior to D. This suggests that in many cases fungi caused considerable yield reduction. However, it should be kept in mind that the toxic effect of D is not restricted to 'toxic cases' and that the average effect in Table 3 is probably also suppressed by phytotoxicity. Fig. 6 indicates, that beet was not influenced by PSS in about 40% of the experiments.

#### *Potato*

PSS increased the yield of potato to 144%/156%. 21 out of 125 experiments were toxic, which is below average. The effect for D was 136%/143%, which was better than for Trapex and Vapam, but less good than for EDB and CP. The averages for heat, Temik and Captan were also high, but this was a result of only one trial. All results together suggest, that control of fungi in addition to nematodes, has been important. Fig. 6 suggests, that PSS increased the yield of potato in a large proportion of the experiments, and that the percentage of soils with increases up to 200% was notable; cf. also 6.2.

#### *Tomato*

The yields for this crop were increased to 120%/126%, therefore less than for potato, and the difference between D and the broad-spectrum soil sterilants was small. EDB was less effective on average. Similarly the percentage of trials without effect or with little effect was relatively high. Extremely high results, therefore failures of the untreated controls, were practically absent. Experiments with tomato were usually laid out in tomato glasshouses, and the results indicate that yield deficiencies on these soils are relatively low, probably owing to earlier PSS treatments as common practice for tomato growing.

#### *Cabbage*

Cabbage reached 128%/144% on the treated soils. 6 out of 22 experiments were toxic cases, especially due to damage by D. A number of soils were apparently problem soils considering the great effects of PSS.

#### *Lettuce*

Lettuce reached 139%/158%, which is a high response. 10 out of 36 experiments were toxic cases, and this is also a high percentage. Fig. 6 suggests, that there was no response in about 40% of the experiments, but that in other fields the response was very high and, therefore, that serious yield deficiencies occurred.

#### *Endive*

For this crop 125%/130% was reached. Yield increase occurred more often than in lettuce, but showed no high figures. Fields with strong yield deficiencies, were therefore absent.

### Carrot

PSS increased the yield of carrot to 118%/129%, which is moderate. 49 out of 190 experiments were toxic cases. This is a high percentage and phytotoxicity may have decreased not only the overall average, but the figure of 129% as well. The effect for D was superior to that for CP and Vapam, and although incidental figures for heat, MB and Trapex were higher than for D, no convincing evidence is available that fungi were playing a major role. Fig. 6 indicates, that many carrot crops were not noticeably influenced.

### Rose

Rose reached 153%/168%. These figures are high, as is the case for most woody crops. These crops were all heavily damaged by *Pratylenchus penetrans* which is widespread in most nursery soils and must have caused high yield depressions in many experiments. D was somewhat more effective than CP, but less than heat, Trapex and other treatments. It is therefore probable that 'specific rose disease', which is attributable to fungi and not to nematodes (OOSTENBRINK and HOESTRA 1961), was present in some of the soils used for our experiments. Analysis of the data, in Fig. 6, shows that the yield of rose is usually improved by PSS, and that often very high percentages, therefore very low yields on untreated soils, occurred.

### Strawberry

PSS increased strawberry yields to 117%/125%. Similar results were obtained with all kinds of sterilizers, and it is one of the few crops in which PCNB was also effective. It is possible that different groups of noxious organisms are involved. Extremely high yield increases are not found (Fig. 6), but it must be kept in mind that the soils used were not typical strawberry soils.

### Apple

PSS increased growth of apple trees to 166%/180%. 58 of the 390 experiments were toxic cases. Most of the experiments were made in orchards on clay soils where 'specific apple replant disease' reduced growth considerably; in others *Pratylenchus penetrans* was present, or also present. The effects of D, Trapex and Vapam were almost similar, but heat and CP were much more effective, whereas MB was intermediary. Such results are already reported by HOESTRA (1968). It is noteworthy that Captan as well compensated part of the yield loss, whereas PCNB did not. The analysis of the results in Fig. 6 shows that the stimulating influence of PSS was present in most of the experiments and that the effects were often drastic.

### Malus

Apple seedlings and rootstocks reached 152%/162%. Analysis of the results shows that the influence of PSS is present in nearly all cases and reaches very high values, as for apple trees. They are nevertheless recorded separately, because apple seedlings and rootstocks are usually grown in nurseries on sandy

soils where the poor growth is often due to nematodes, especially *P. penetrans*. This is substantiated by the fact that D, Trapex and Vapam were not inferior to CP and MB for these plants.

#### *Pear and Prunus*

These plants were usually damaged in the test soils by *P. penetrans* and reached high yield increases, namely 143%/147% and 185%/194% respectively. D was, correspondingly, as effective or better than CP, MB, Trapex or Vapam.

#### *Chrysanthemum*

PSS increased the yield of chrysanthemum to 115%/119%, with few toxic cases. The effect is moderate, which is explained by the fact that most data on chrysanthemum are from soils where the plant was never grown before and where the soil flora and fauna obviously contained few noxious species. The few high scores, recorded in Fig. 6, are from fields where high densities of noxious nematodes, amongst others *P. penetrans*, were present.

#### *Narcissus*

The effect for narcissus was 121%/123%, with only 1 toxic case amongst 35 experiments. D, EDB, Trapex and Vapam were about equally effective. The result was probably mainly due to the elimination of parasitic nematodes, especially *P. penetrans* which is the cause of root rot. Fig. 6 shows, that there were few experiments without a positive response, but also that the response was always moderate, as could be expected from the fact that PSS is an established technique in narcissus culture on the soils used for our experiments. Crop failures, and correspondingly strong response to PSS, do not occur under these circumstances.

#### *Iris*

PSS increased the yield of iris to 125%/132%. This was mainly due to the strong effect of Vapam, may be because special fungi were controlled. At any rate Vapam was more effective than D in this case. Fig. 6 suggests, that there will be fairly high yield losses for iris on many soils when not treated.

#### *Gladiolus*

Gladiolus reached only 109%/115%, and this was due to the effect of D in particular which reached 118%/118% and was superior to Trapex and Vapam. The effect was probably mainly nematode control.

#### *Hyacinth*

Hyacinth also reached only 111%/115%. For this crop Vapam and Trapex were applied, probably for the control of fungi and bacteria. The problems in hyacinth and iris are related and different from those in narcissus and gladiolus, considering the response to different soil disinfectants.

### *Other crops*

The other crops reached the high percentage of 168%/191%. This was mainly due to the fact that many of these crops were woody plants which were tested on soils infested with *P. penetrans* and other nematodes. It is, for this reason, understandable, that D was as effective or better than any of the other treatments, including CP, MB, Trapex and Vapam. Fig. 6 shows that many experiments indicate yield deficiencies, which may be very high as in the case of malus, pear and prunus.

Comparison of the effect on different crops reveals, that PSS did not cause notable effect (grey columns in the diagrams of Fig. 6) in more than half or about half of the experiments for zinnia, chrysanthemum, hyacinth, oat, tomato, lettuce and carrot. These crops were not responsive in quite so many cases for different reasons: zinnia and chrysanthemum because these plants were often tested in fresh soils. Hyacinth and tomato because they were grown in soils where PSS was already common practice. Lettuce and carrot because these crops as well were often tested in fresh soils (whereas other soils showed great yield deficiencies and therefore great yield response to PSS – these certainly are crops which do not maintain high productivity if grown intensively without application of PSS), and finally oat which apparently is less affected by soil-borne yield deficiencies than any other agricultural crop, although great yield losses indicated by PSS effects occur in a number of cases.

Pea, barley, and woody crops (pear, apple, malus, prunus, and also rose and the other crops, which were mostly woody plants) seldom reached a normal yield unless PSS was applied. In practice: pea and barley are known as risky crops, noxious nematode infestations are widespread, and it is known for pea and suspected for barley that the same is true for pathogenic soil fungi. The woody crops usually suffer much from specific replant diseases (in the case of orchard trees) and from nematode infestations (in the case of nursery plants).

Intermediary in their response are important crops as grass, clover, potato and cabbage, for which PSS causes rewarding yield increases on many soils. This refers, of course, to yield improvements and does not evaluate economic rentability of the treatments.

#### *4.3.4. PSS results related to the presence of parasitic organisms*

Table 5 lists the experimental results in categories according to the presence of noxious pests or diseases in the soil, with a separate category for 'No pathogenic nematodes present' as well as for 'No infestation known' which means that the experiments were laid on normal fields without known problems or known infections at the moment of selection. The categories are placed in order of decreasing PSS-effects, and separate 'No infestation known' and 'Total'. Some conclusions can be made, despite the fact that the indication of some dominant pathogen does not represent the hygienic condition of a soil for all crops. The categories will be discussed separately.

Table 6 and 7 are specifications of Table 5.

TABLE 5. Computed average yield percentage on treated soils (untreated = 100), separately for soils with different parasitic organisms. Between brackets are the numbers of experimental items on which the average percentages are based.

Parasitic organism or disease	General average	Average without toxic cases
1 <i>Heterodera</i> spp.	547 (6)	638 (5)
2 <i>H. rostochiensis</i>	190 (43)	208 (37)
3 Specific apple replant disease	173 (348)	185 (305)
4 <i>Pratylenchus penetrans</i>	168 (589)	185 (495)
5 <i>P. crenatus</i> , <i>Tylenchorhynchus dubius</i> and <i>Rotylenchus robustus</i>	141 (190)	154 (155)
6 <i>R. robustus</i>	132 (18)	139 (16)
7 <i>Hemicycliophora</i> and <i>Paratylenchus</i> spp.	126 (123)	135 (98)
8 <i>Meloidogyne</i> spp.	120 (166)	126 (140)
9 <i>Pythium</i> spp.	111 (196)	115 (163)
10 No pathogenic nematodes present	99 (33)	114 (17)
11 No infestation known	115 (741)	125 (541)
12 Total	141 (2453)	154 (1972)

They furnish the reactions of special crops on soils with a certain parasite, and therefore comprise information on specific host-parasite relationships.

#### *Heterodera* spp.

There were only 6 experiments, 5 on pea in a soil with *H. goettingiana* and 1 on clover in a soil with *H. trifolii*, and generalization of the data is therefore not justified. The very high yield increase, 547%/638%, was only due to the severe damage which pea suffered in the heavily infested soil. Concerning *H. rostochiensis*, the yield increase obtained reflects the great sensitivity to nematicides of this nematode of potato and tomato, the only plants which were tested on soil with known infestation. If, within this category, the growth of potato and tomato is rated separately, the figures become 198%/223% and 154%/154%, respectively.

#### *Specific apple replant disease (SARD)*

Results only for apple within this category would have been 175%/188%. Soils with SARD, however, usually comprise *P. penetrans* as well. It was not possible to separate the effects of the factors involved in the figures. The fact that pear, chrysanthemum and 'other crops' also reached high yield increases makes it doubtful whether this group of soils was defined more by SARD than by *P. penetrans*.

#### *Pratylenchus penetrans*

The figures for different crops within the group vary greatly. Considering the results based on more than 5 experiments we found that in this category prunus, rose, apple trees, malus seedlings, black-currant and 'other crops' (most

TABLE 6. Computed average yield percentage on treated soils (untreated = 100), separately for soils with different parasitic organisms. The column numbers indicate the parasitic organisms or diseases found; cf. Table 5. Between brackets are the number of experimental items on which the percentage is based.

Parasitic organisms; cf. Table 5.												
Crops	1	2	3	4	5	6	7	8	9	10	11	12
Oat	-	(-)	-	101 (6)	237 (9)	-	(-)	-	(-)	-	110 (30)	134 (45)
Wheat	-	(-)	-	107 (3)	114 (1)	-	122 (11)	-	(-)	-	-	119 (15)
Barley	-	(-)	-	103 (4)	145 (1)	-	(-)	131 (10)	(-)	-	120 (1)	124 (16)
Grass	-	(-)	-	137 (7)	153 (14)	-	145 (16)	-	(-)	102 (10)	114 (40)	126 (87)
Clover	93 (1)	(-)	-	122 (13)	202 (6)	-	(-)	-	(-)	-	106 (33)	121 (53)
Pea	738 (5)	(-)	-	91 (6)	141 (7)	-	(-)	-	(-)	-	123 (1)	255 (19)
Bean	-	(-)	-	84 (7)	131 (12)	-	(-)	-	(-)	-	-	114 (19)
Beet	-	(-)	-	120 (16)	107 (11)	-	202 (19)	-	(-)	-	110 (35)	133 (81)
Potato	-	198 (35)	-	115 (23)	138 (12)	-	106 (11)	-	(-)	102 (3)	130 (41)	144 (125)
Tomato	-	154 (8)	-	157 (4)	49 (1)	-	(-)	118 (147)	(-)	-	75 (1)	120 (161)
Cabbage	-	(-)	-	118 (17)	-	-	(-)	159 (5)	(-)	-	-	128 (22)
Lettuce	-	(-)	-	130 (2)	-	-	(-)	-	(-)	-	139 (34)	139 (36)
Endive	-	(-)	-	133 (4)	-	116 (4)	-	108 (4)	(-)	-	129 (15)	125 (27)
Carrot	-	(-)	-	120 (4)	136 (28)	137 (14)	106 (11)	-	(-)	96 (10)	115 (123)	118 (190)
Rose	-	(-)	-	189 (93)	138 (16)	-	(-)	-	(-)	99 (10)	104 (54)	153 (173)
Strawberry	-	(-)	-	139 (3)	108 (22)	-	(-)	-	(-)	-	119 (71)	117 (96)
Apple	-	(-)	175 (318)	183 (18)	-	-	(-)	-	(-)	-	109 (54)	166 (390)
Malus	-	(-)	-	158 (93)	130 (22)	-	(-)	-	(-)	-	116 (3)	152 (118)
Pear	-	(-)	140 (24)	164 (3)	-	-	(-)	-	(-)	-	-	143 (27)
Prunus	-	(-)	-	204 (42)	145 (6)	-	(-)	-	(-)	-	120 (9)	185 (57)
Black-currant	-	(-)	-	149 (6)	126 (6)	-	(-)	-	(-)	-	93 (19)	110 (31)
Liguster	-	(-)	-	117 (23)	-	-	(-)	-	(-)	0	103 (24)	110 (47)
Chrysanthemum	-	(-)	199 (2)	-	-	-	(-)	-	(-)	-	109 (30)	115 (32)
Zinnia	-	(-)	-	-	-	-	(-)	-	(-)	-	103 (21)	103 (21)
Narcissus	-	(-)	-	123 (35)	-	-	(-)	-	(-)	-	-	123 (35)
Iris	-	(-)	-	-	-	-	100 (11)	-	(-)	-	140 (19)	125 (30)
Gladiolus	-	(-)	-	-	-	-	108 (11)	-	(-)	-	111 (1)	109 (12)
Hyacinth	-	(-)	-	-	-	-	-	-	111 (196)	-	-	111 (196)
Other crops	-	(-)	215 (4)	207 (157)	167 (16)	-	103 (33)	-	(-)	-	117 (82)	168 (292)
Total	547 (6)	190 (43)	173 (348)	168 (589)	141 (190)	132 (18)	126 (123)	120 (166)	111 (196)	99 (33)	115 (741)	141 (2453)

TABLE 7. Computed average yield (excluded toxic cases) percentage on treated soils (untreated = 100), separately for soils with different parasitic organisms. The column numbers indicate the parasitic organisms or diseases found; cf. Table 5. Between brackets are the number of experimental items on which the percentage is based.

Parasitic organisms cf. Table 5.												
Crops	1	2	3	4	5	6	7	8	9	10	11	12
Oat	- (-)	- (-)	- (-)	104 (4)	237 (9)	- (-)	- (-)	- (-)	- (-)	- (-)	116 (23)	145 (36)
Wheat	- (-)	- (-)	- (-)	131 (1)	114 (1)	- (-)	124 (10)	- (-)	- (-)	- (-)	- (-)	124 (12)
Barley	- (-)	- (-)	- (-)	109 (3)	145 (1)	- (-)	- (-)	171 (6)	- (-)	- (-)	120 (1)	147 (11)
Grass	- (-)	- (-)	- (-)	137 (7)	153 (14)	- (-)	149 (15)	- (-)	- (-)	105 (7)	119 (34)	131 (77)
Clover	- (-)	- (-)	- (-)	138 (9)	202 (6)	- (-)	- (-)	- (-)	- (-)	- (-)	115 (21)	135 (36)
Pea	638 (5)	- (-)	- (-)	155 (2)	158 (6)	- (-)	- (-)	- (-)	- (-)	- (-)	123 (1)	326 (14)
Bean	- (-)	- (-)	- (-)	122 (2)	139 (10)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	136 (12)
Beet	- (-)	- (-)	- (-)	178 (7)	125 (7)	- (-)	225 (16)	- (-)	- (-)	- (-)	127 (19)	166 (49)
Potato	- (-)	223 (29)	- (-)	122 (18)	138 (12)	- (-)	108 (9)	- (-)	- (-)	109 (2)	138 (34)	156 (104)
Tomato	- (-)	154 (8)	- (-)	192 (3)	- (-)	- (-)	- (-)	122 (126)	- (-)	- (-)	- (-)	126 (137)
Cabbage	- (-)	- (-)	- (-)	133 (12)	- (-)	- (-)	- (-)	177 (4)	- (-)	- (-)	- (-)	144 (16)
Lettuce	- (-)	- (-)	- (-)	130 (2)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	160 (24)	158 (26)
Endive	- (-)	- (-)	- (-)	151 (3)	- (-)	122 (3)	- (-)	108 (4)	- (-)	- (-)	133 (14)	130 (24)
Carrot	- (-)	- (-)	- (-)	120 (4)	152 (21)	143 (13)	106 (11)	- (-)	- (-)	112 (2)	126 (90)	129 (141)
Rose	- (-)	- (-)	- (-)	194 (89)	141 (15)	- (-)	- (-)	- (-)	- (-)	126 (6)	116 (31)	168 (141)
Strawberry	- (-)	- (-)	- (-)	139 (3)	115 (15)	- (-)	- (-)	- (-)	- (-)	- (-)	127 (57)	125 (75)
Apple	- (-)	- (-)	188 (277)	183 (18)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	120 (37)	180 (332)
Malus	- (-)	- (-)	- (-)	158 (82)	130 (18)	- (-)	- (-)	- (-)	- (-)	- (-)	125 (2)	162 (102)
Pear	- (-)	- (-)	144 (22)	164 (3)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	147 (25)
Prunus	- (-)	- (-)	- (-)	204 (42)	253 (2)	- (-)	- (-)	- (-)	- (-)	- (-)	125 (8)	194 (52)
Black-currant	- (-)	- (-)	- (-)	149 (6)	137 (5)	- (-)	- (-)	- (-)	- (-)	- (-)	113 (6)	133 (17)
Liguster	- (-)	- (-)	- (-)	137 (14)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	115 (15)	125 (29)
Chrysanthemum	- (-)	- (-)	199 (2)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	113 (25)	119 (27)
Zinnia	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	109 (13)	109 (13)
Narcissus	- (-)	- (-)	- (-)	124 (34)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	123 (34)
Iris	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	104 (8)	- (-)	- (-)	- (-)	144 (18)	132 (26)
Gladiolus	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	115 (8)	- (-)	- (-)	- (-)	111 (1)	115 (9)
Hyacinth	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	- (-)	115 (163)	- (-)	- (-)	115 (163)
Other crops	- (-)	- (-)	215 (4)	238 (127)	189 (13)	- (-)	108 (21)	- (-)	- (-)	- (-)	125 (67)	191 (232)
Total	638 (5)	208 (37)	185 (305)	185 (495)	154 (155)	139 (16)	135 (98)	126 (140)	115 (163)	114 (17)	125 (541)	154 (1972)



of which were woody plants) were greatly promoted by PSS. Grass, clover and narcissus were promoted moderately, whereas oat and barley were hardly influenced. These results agree with earlier knowledge about the significance of *P. penetrans*. The fact that potato yields reached only 115%/122%, is probably due to the fact that the infestation rate of *P. penetrans* on most of the test soils was low, for potato is a suitable host and is susceptible to damage by high *P. penetrans* densities. Beet is known as an unsuitable host, but it reached a higher yield increase than potato, viz. 120%/178%. The last figure is based on 7 experiments only. Scrutiny of the incidental results of the basic list indicates that the applied chemical doses in 11 out of the 12 potato experiments varied between low to moderately low, which agrees with the conclusions concerning chemical doses (cf. 4.3.2. and Fig. 3).

#### *P. crenatus, Tylenchorhynchus dubius and Rotylenchus robustus*

Significant regressions for the yield of some gramineous crops on the density of this complex were demonstrated, whereas beet, potato and other crops are on record to be less susceptible. This agrees in general with our results. Considering the results for the crops with more than 5 experiments within this category we find a low response for beet, and a high response for oat, clover and grass, and also for pea and carrot if the toxic cases are excluded (cf. Table 7). Bean, potato and some woody crops, however, are also fairly responsive on these soils. This was unexpected, at least for potatoes. The incidental trial results indicate that the effect for oat was due to a synergistic role of plant-parasitic nematodes in the presence of other soil micro-organisms (cf. 6.9 and 8).

#### *R. robustus*

The main crop in this category was carrot, of which the results obtained were similar to those in the preceding category.

#### *Hemicycliophora and Paratylenchus spp.*

The effect reached was 126%/135%, but for some susceptible crops much higher figures were reached, e.g. 202%/225% for beet in soils with *Hemicycliophora* spp. and 145%/149% for grass (in soils with *Paratylenchus* spp. or with *Hemicycliophora* spp. Scrutiny of the incidental results of the basic list indicates a considerable effect of the broad-spectrum chemical CP and fungicide Captan in beet experiments due to control of specific beet fungi regardless of *Paratylenchus* spp. A fairly moderate response for the other chemicals was also obtained by other crops. The response due to the specific nematicidal treatments when *Hemicycliophora* spp. was present was greater on average than when *Paratylenchus* spp. was present. This indicates the greater aggressiveness of the former against the latter pest; in spite of the suitability of a wider variety of pathogenic fungi to survive with the second one in its wet ecological niche.

#### *Meloidogyne spp.*

The yield increase in experiments with *Medoidogyne* spp. reached only

120%/126%, despite the fact that they are known to be powerful parasites. The damage they cause is, however, mainly restricted to glasshouse crops and is reasonably well under control. 147 of the 166 experiments in this category were with tomato and the figures for this crop were 118%/122%. Much higher figures were reached for barley in soils with *M. naasi*, viz. 130%/171%.

#### *Pythium spp.*

*Pythium* spp. were only studied in soils used for hyacinth cultures, and here the infestation is usually well under control. This explains that the effect was only 111%/115%.

#### *No pathogenic nematodes present*

In these soils hardly any yield increase was obtained with PSS; the average figure from 33 experiments was 99%/114%. They were, however, mainly new polder soils or clean sand or peat in which no pathogenic complexes had built up yet.

The effects obtained were 102%/105% for grass, 102%/109% for potato, 96%/112% for carrot and 99%/126% for rose.

#### *No infestation known*

The category for unknown cases is interesting, because it is in fact the opposite of the categories selected for the presence of problems or pathogens. The effect for this category is 115%/125%. This is relatively low, partly because most of these soils were included in a series of experiments where, regrettably, unusual phytotoxicity was encountered, and this must also have depressed the figure of 125%. At least 125% is therefore an attainable average for soils considered to be free from special problems.

Table 7, which excludes the toxic cases, is useful to search for hidden problems which can be solved by PSS. Crops with relatively high yield increases in the category 'No infestation known', and for this reason possibly comprising hidden problems are lettuce, iris, potato, endive, beet, strawberry and carrot. Some of these problems will be specially studied in the following chapters.

#### *4.3.5. PSS results in relation to soil type*

Table 8 lists the experimental results according to the corresponding soil type. It is well known that the occurrence of diseases and pests is correlated with soil type.

By PSS high yield figures were reached on loamy sand (185%/198%). The percentage of toxic cases was very low, namely 10%, but the degree of toxicity was very high. The effects on clay soil and on peaty sand were somewhat less (158%/174% and 156%/174% respectively), but they were higher than on diluvial sand and on dune sand. The effect on silty soil is exceedingly low, namely 113%/119%. This is also true for polder sand, which reached 111%/127%, but in this case many experiments were often laid on new, uninfested polder soil. PSS in the few experiments with river sand caused hardly any

TABLE 8. Computed average yield percentage on treated soils (untreated = 100) separately for different types of soil. Between brackets are the numbers of experimental items on which the percentage is based.

Type of soil	General average	Average without toxic cases	Average for toxic cases only
Loamy sand	185 (62)	198 (56)	60 (6)
Clay	158 (459)	174 (376)	87 (83)
Peaty sand	156 (768)	174 (620)	82 (148)
Diluvial sand	125 (503)	136 (389)	86 (114)
Dune sand	120 (311)	126 (262)	90 (49)
Silt	113 (229)	119 (181)	90 (48)
Polder sand	111 (28)	127 (17)	85 (11)
River sand	94 (15)	108 (7)	82 (8)
Other and unknown	134 (77)	144 (63)	90 (14)
Total	141 (2453)	154 (1972)	86 (481)

growth improvement, which could be expected because this soil does not comprise plant pathogens and harbours few other organisms. It is well known that many treatments were toxic in this soil due to the limitation of soil particles surface and accordingly high chemical concentration. It must be concluded that the effect of PSS varies greatly between soils. The strong effects for clay and peaty sand compared to diluvial or dune sand were not expected. The fact that PSS is not popular for clay soils must be due to difficult application and aeration. The high effect for loamy sand and the low effect for silt are also noteworthy.

#### 4.3.6. PSS results in relation to locations

Listing of the results according to the PD districts mentioned in Table 9 is arbitrary. It nevertheless reveals or confirms some general conclusions. It confirms that the effect of PSS differs greatly between localities.

The high figures obtained in Winschoten and Wageningen may be related to soil type (cf. 4.3.5.), but they are no doubt also biased by the facts that: a. many experiments in Winschoten were made on soils infested with *P. penetrans* and: b. that in Wageningen several infested soils were brought together.

The very low figure for Emmeloord (107%/112%) reflects the fact that many experiments were made on new, uninfested polder soils. The results for Lisse, 115%/116%, probably reflect the situation in the bulb soils where diseases are already well under control and where PSS is an established technique, considering also the very low percentage of toxic cases. Further analysis of the data would result in repetition of the conclusions recorded earlier and is therefore omitted.

TABLE 9. Computed average yield percentage (untreated = 100) separately for the experiments made in different locations. Between brackets are the number of experimental items on which the percentages are based.

Locations	General average	Average without toxic cases	Average for toxic cases only
Winschoten	164 (577)	186 (459)	79 (118)
Wageningen	161 (238)	180 (191)	85 (47)
Zwolle	126 (70)	134 (58)	87 (12)
Leiden	124 (108)	138 (78)	87 (30)
Utrecht	123 (117)	129 (99)	91 (18)
Rotterdam	121 (71)	129 (58)	83 (13)
Almelo	118 (53)	125 (43)	87 (10)
Lisse	115 (120)	116 (112)	96 (8)
Exloo	112 (54)	121 (37)	91 (17)
Deventer	112 (93)	124 (60)	90 (33)
Emmeloord	107 (149)	112 (108)	93 (41)
Other locations	130 (803)	139 (669)	86 (134)
Total	141 (2453)	154 (1972)	86 (481)

## 5. PSS ON FIELDS WITHOUT KNOWN PROBLEMS

### 5.1. INTRODUCTION

The inventory mentioned under 4. comprised all data available from field experiments complemented with few data from experiments in pots.

Most of the fields and soils used were selected for the presence of problems or parasites, although the data from experiments from soils with no known problems were also included. The results at any rate must be biased by the choice of the fields, and the average effect for all soils must be lower than the effect found in our inventory.

However the inventory also comprised a series of results with crops on 11 different soils well distributed all over the country. They were selected for the absence of problems or known parasites and on them extensive experiments with 3 replicates of different treatments were made.

Each soil obtained three different treatments, according to a standardized scheme, with the broad-spectrum disinfectant chloropicrin (= CP), the nematocide dichloropropene (= D), the fungicide pentachloronitrobenzene (= PCNB) and untreated as control (= C). The result of this series of experiments is to a certain extent representative, but is contrary to the general inventory with a bias to problem-free soil. The average result is therefore lower than would be expected as average for all soils in the country. The series of treatments were applied in autumn 1966 and spring 1967 and, unexpectedly, showed an exceptionally high number of toxic cases among the test crops in 1967. The average results for this reason are also below an overall average. The low applied chemical doses are contributing also in lowering the results (cf. 4.3.2 & Fig. 3). They may nevertheless be useful to indicate the minimum limit of PSS effects in Dutch soils, and have therefore been lifted from the basic list in Chapter 4. and presented in detail hereafter.

### 5.2. EXPERIMENTS

The work was realized in cooperation with the local offices of the PD. The 11 fields are numbered and described hereafter, with indications about the data of treatment and the data of sowing or planting: (see page 52).

The treatments applied were as follows:

- D = dichloropropene, in the formulation DD at the rate of 25–40 ml/m<sup>2</sup>;
- CP = chloropicrin at the rate of 25–40 ml/m<sup>2</sup>;
- PCNB = pentachloronitrobenzene in the formulation Brassicol at the rate of 20 g/m<sup>2</sup>;
- C = untreated control.

<i>Number and PD district</i>	<i>Soil type</i>	<i>Treatment date</i>	<i>Sowing or planting date</i>
1. Almelo	Peaty sand	8.11.1966	13.4.1967
2. Assen	Diluvial sand	21.10.1966	14.4.1967
3. Den Bosch	Diluvial sand	7.11.1966	7.4.1967
4. Emmeloord	Light clay	27.10.1966	27.4.1967
5. Exloo	Peaty sand	8.11.1966	21.4.1967
6. Groningen	Diluvial sand	20.4.1967	23.5.1967
7. Leeuwarden	Silty clay	20.4.1967	16.5.1967
8. Leiden	Silty clay	20.4.1967	10.5.1967
9. Utrecht	Light clay	19.4.1967	11.5.1967
10. Winschoten	Peaty sand	9.11.1966	25.4.1967
11. Zwolle	Diluvial sand	8.11.1966	12.4.1967

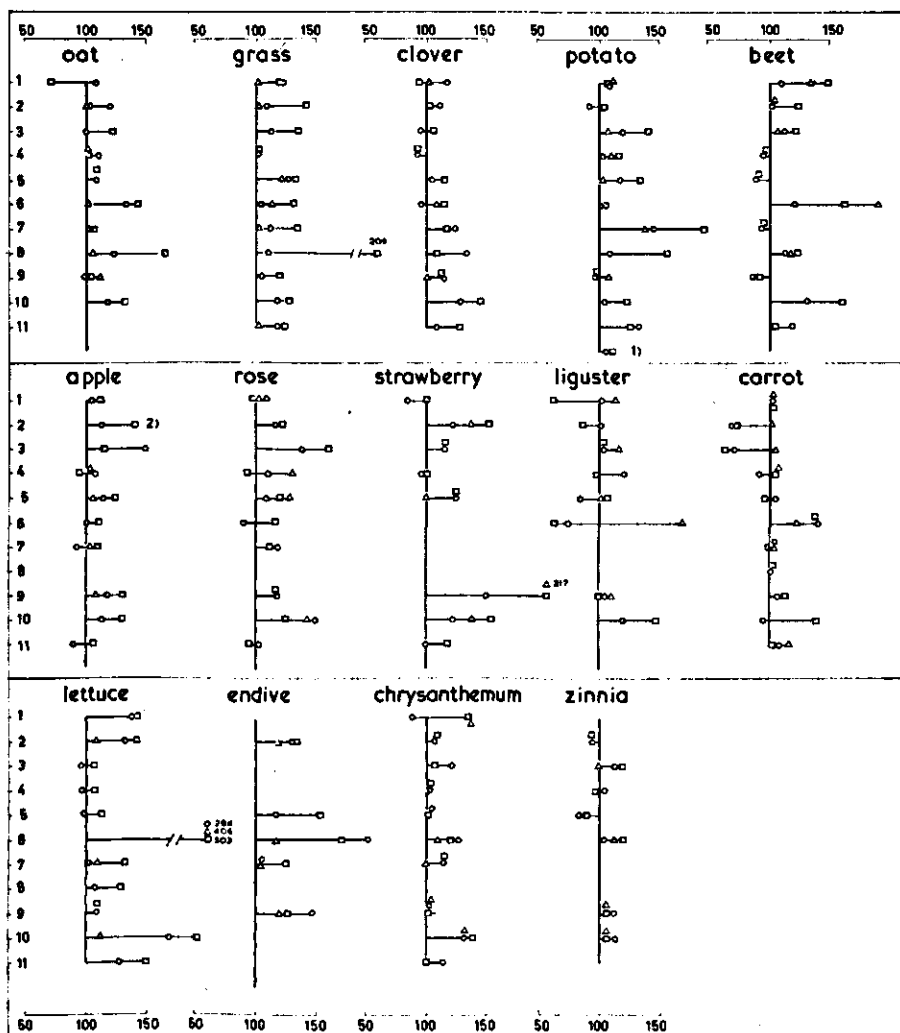
The crops grown were oat, grass, clover, potato, beet, apple, rose, strawberry, liguster, carrot, lettuce, endive, chrysanthemum, zinnia.

Not alle crops were present on all fields (cf. Fig. 7).

The results are calculated as percentage of control in the same way as in chapter 4. They are represented in Table 10 and Fig. 7. In Fig. 8 development of some crops on differently treated soils is recorded with time.

TABLE 10. Soils without known problems. Calculated average yield percentages on treated soil (untreated = 100) for all replicates of 14 tested crops with and without toxic cases and indication to the most favourite PSS used and its general average. Between brackets the number of experiments carried out.

Test crops	General average	Average without toxic cases	PSS treatment which was on average most effective	Average of best PSS treatments
Oat	109 (30)	114 (23)	CP	123 (10)
Grass	116 (33)	119 (29)	CP	134 (11)
Clover	105 (33)	114 (20)	D	117 (11)
Potato	113 (36)	119 (28)	CP	126 (12)
Beet	112 (33)	126 (19)	CP	124 (11)
Apple	108 (30)	115 (21)	CP	122 (10)
Rose	110 (30)	122 (20)	CP	125 (10)
Strawberry	123 (24)	135 (18)	CP	138 (8)
Liguster	103 (24)	115 (15)	PCNB	123 (8)
Carrot	102 (33)	111 (21)	CP	112 (11)
Lettuce	140 (33)	162 (23)	CP	182 (11)
Endive	129 (15)	133 (14)	CP	151 (5)
Chrysanthemum	109 (30)	112 (25)	CP	115 (10)
Zinnia	100 (24)	109 (13)	D	106 (8)
Total	113 (408)	122 (289)		128 (136)



<sup>1</sup> An extra field at Dronten was planted with potato only.

<sup>2</sup> Apple seedlings instead of trees.

FIG. 7. Influence of chloropicrin (□), DD (○) and pentachloronitrobenzene (Δ) on yield (percentage of untreated control) of 14 crops in 11 trial fields without known parasites chosen at random all over The Netherlands. Pentachloronitrobenzene was toxic in most cases so for this chemical only results of non-toxic items are drawn. Abscissa for each crop graph: percentage yield comparable to check. Ordinate for each crop graph: numbers to indicate trial fields, cf. 5.2.

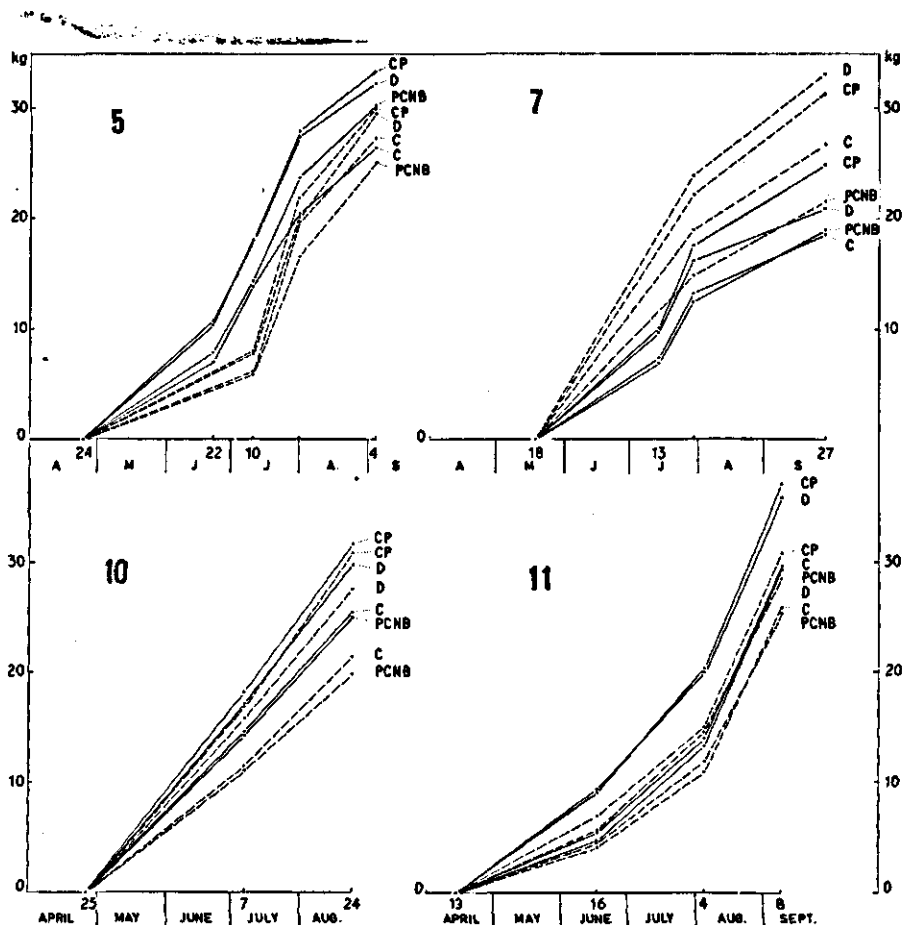


FIG. 8. Yield of grass (—) and of clover (---), estimated from successive cuts for four representative trial fields, namely at Exloo (nr. 5), Leeuwarden (nr. 7), Winschoten (nr. 10) and Zwolle (nr. 11), as influenced by PSS with D, CP and PCNB compared to the untreated C. Abscissa for each field: time scale with planting and cutting dates specially recorded. Ordinate for each field: yield in kilograms per 2 m<sup>2</sup> as average of 3 replicates, cumulative.

### 5.3. DISCUSSION

Table 10 indicates an average yield for alle crops on all treated soils of 113% (based on 408 items), or 122% if the toxic cases are omitted (289 items), or 128% for the average best treatment of each crop (136 items). The average is 156% for the best treatments of each soil-crop combination (42 items each based on 3 replicates). The last mentioned result can therefore be reached on random fields without recognised problems by proper choice of the soil-crop treatments. The potential effect of PSS on all agricultural land including the problem fields must be higher than 156%.



Comparison of the treatments reveals that PCNB has caused phytotoxicity in many cases; these have not been drawn in Fig. 7. For some crops on some soils, however, it has caused notable growth increases, namely on soil 1 for beet, on soil 2 for strawberry, on soils 4 and 5 for rose, on soil 6 for beet, liguster, carrot and lettuce, on soil 7 for potato, on soil 9 for strawberry, and on soil 10 for rose, strawberry and chrysanthemum. PCNB was as good or better than CP or D only for rose, strawberry and liguster in a few fields, presumably because noxious pathogenic fungi were controlled.

Of the other crops the treatment with CP was generally more effective than D and less toxic. This was on average true for individual crops, except for clover and zinnia which were promoted more by D (Table 10).

Fig. 7, however, shows that individual crop-soil combinations often yielded other results. CP was considerably less effective than D in field 1 for oat, clover and liguster, in field 2 for liguster, in field 3 for apple and chrysanthemum, and in field 4 for apple, rose and liguster. CP was also markedly phytotoxic in most of these as well as in some other cases, namely in field 1 for oat, clover and liguster, in field 2 for liguster, in field 3 for carrot, in field 4 for clover, apple and rose, in field 5 for beet and zinnia, in field 5 for liguster, and in field 9 for beet. D caused more toxic cases than CP, but the most severe cases of phytotoxicity are due to CP (oat in field 1, liguster in fields 1 and 6, carrot in field 3). The marked phytotoxicity by CP in this series of experiments is unusual, and it must have depressed the overall results.

Table 10 shows that all crops are invigorated by the CP and D treatments on most soils, but to different degrees. High responses for the generally best treatment are obtained for lettuce, endive, strawberry, grass, potato, rose and beet, whereas the score for zinnia, carrot and chrysanthemum is low. High average scores in some of the fields are often exceedingly strong responses, therefore low control yields. The results of Fig. 7 demonstrate that nearly all crops suffered heavy yield losses on some fields, and that nearly all fields harboured noxious problems for some crops. This strongly varied pattern probably reflects the general situation in practical fields. Most of these strong responses or yield deficiencies require detailed diagnostic study to unravel the cause or causes of the deficiency.

Fig. 8 illustrates plant development on treated and untreated soil with time. The graphs recorded are for grass and for clover on fields 5, 7, 10 and 11, but they represent the usual pattern. It appears to be common that differences in yield between treatments which occur shortly after sowing or planting, are maintained and increase with time during the growing season.

## 6. PSS OF SOILS WITH KNOWN NEMATODE PESTS

### 6.1. INTRODUCTION

Five selected PSS treatments to study their effects on growth of a susceptible host plant, were applied to eight soils infested with a known noxious nematode. All soils were treated in open bins in the laboratory and perspex tubes of 350 g were then filled with it. There were 10 replicates per treatment. Three weeks after treatment the pots were sown or planted and placed in a lighted glasshouse heated to between 15 and 25°C; the plants received water and fertilizer of a standard Hoagland solution (cf. 7.1.) at the rate of 10 ml/500 ml soil, according to the apparent need of the plants without differences between the objects in an experiment. The plants were extensively evaluated after 10–14 weeks; nematode populations were enumerated before and after the experiment.

The treatments were:

- a. H = heat, viz. 60°C for 2 hours applied in flat, metal containers sunk in a water bath;
- b. CP = chloropicrine, 0.3 ml per l soil (comparable to 60 ml/m<sup>2</sup> in the field), applied to the soil in a plastic bag placed in a closed bin;
- c. D = dichloropropene in the formulation DD, 0.15 ml per 1 of soil (comparable to 30 ml/m<sup>2</sup> in the field), applied in the same way as CP;
- d. T = Temik, 0.025 g per 1 of soil (comparable to 5 g/m<sup>2</sup> in the field);
- e. CA = Captan, 0.3 g per 1 of soil (comparable to 60 g/m<sup>2</sup> in the field);
- f. C = check, i.e. untreated soil.

The soils used, the nematodes present and the plants grown were as follows:

- a. Sandy soil from Wageningen with *Heterodera rostochiensis* Woll. grown with potato, *Solanum tuberosum* L. 'Deodara';
- b. Clay soil with *Heterodera goettingiana* Liebscher grown with pea, *Pisum sativum* L. 'Rovar';
- c. Polder sand from the Wieringermeer with *Hemicycliophora conida* Thorne grown with ryegrass, *Lolium perenne* L. 'Orig. Weide type';
- d. Sandy soil with a monoculture of *Meloidogyne javanica* (Treub) grown with tomato *Lycopersicum esculentum* L. 'Moneymaker' in the glasshouse of the PD;
- e. Polder-clay soil from Oostelijk Flevoland with *Meloidogyne naasi* Franklin grown with cabbage, *Brassica napus* L. var. *napus* 'Lihonova';
- f. Polder-clay from Oostelijk Flevoland infested with *M. naasi* grown with barley, *Hordeum vulgare* L. 'Delisa';
- g. Sandy polder soil from Middenmeer, Oostelijk Flevoland, infested with *Meloidogyne naasi* and grown with barley;
- h. Loamy sand from the PD garden infested with a complex of *Pratylenchus crenatus* Loof + *Tylenchorhynchus dubius* (Bütschli) + *Rotylenchus robustus* (de Man) and grown with oat, *Avena sativa* L. 'Marne'.

## 6.2. SANDY SOIL WITH *H. ROSTOCHIENSIS* UNDER POTATO

The heavily infested field soil used was kept in a concrete cylinder in the PD garden and had been grown with potato annually for more than 10 years.

The direct effect of the PSS treatments on the nematode populations was measured by determining the active nematodes including *Heterodera* larvae and males in the soils just after the treatment and again at the end of the experiment after the potato test plants had been grown (Table 11). The effects on growth of the potato and on root infestation by *H. rostochiensis* are extensively recorded in Table 12, whereas Figs. 9, 10 and 11 illustrate the main results.

Table 12 and Fig. 9 show that all treatments increased potato growth considerably, without changing the percentage root weight and the percentage dry matter. The plant response was significant for total plant weight, shoot weight, shoot size, root weight, root size as well as feeder roots and root colour. The treatments, however, differed considerably. CA was significantly less effective and CP significantly more so than H and D for most of the statistics used, whereas T was mostly intermediate between CP and H or D, but showed specific deviations which require further consideration. It is noteworthy that T obviously suppresses the number of males and that CA increases it as compared to C.

The nematode data indicate that the plant response is largely, but not in all details, due to the elimination of *H. rostochiensis*. This cannot be concluded from Table 11, which is incomplete, because only the active stages of *Heterodera* in the soil were determined.

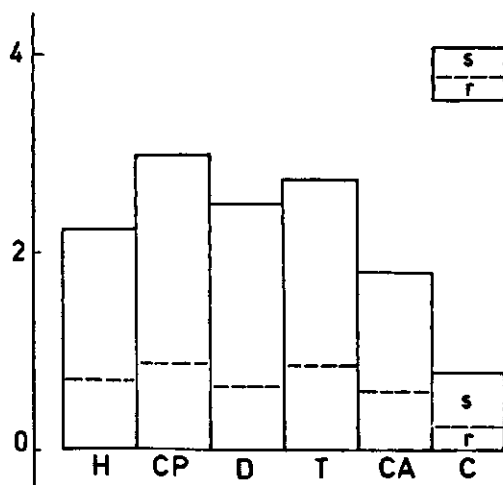


FIG. 9. Effect of different PSS treatments (H, CP, D, T, CA and C; cf. text) on total weight of potato when it was grown in tubes containing 350 g of soil with *H. rostochiensis*. All results are averages of 10 replicate tubes. The least significant differences for r and s are added at the right top corner. Abscissa: PSS treatments as indicated. Ordinate: plant weights in g subdivided in root weight (r) and shoot weight (s).

TABLE 11. Sandy soil with *H. rostochiensis*. Effect of 5 PSS treatments (H, CP, D, T, CA and C; cf. text) on active nematodes in the soil between treatment and planting ( $P_i$ ) and 101 days after planting potato, i.e. on 9.4.1970 ( $P_f$ ). Nematode numbers are per tube of 350 g of soil, as average of 10 replicates. H. l. + ♂ = free *Heterodera* larvae + males, O. = other stylet-bearing nematodes, S. = saprozoic nematodes.

PSS treatments	Nematodes	$P_i$			$P_f$		
	$H.1. + \delta^1)$	$O.$	$S.$	$H.1. + \delta^1)$	$O.$	$S.$	
H	0 + 0	0	0	0 + 0	0	27400	
CP	0 + 0	0	0	0 + 0	0	15400	
D	0 + 0	0	0	0 + 0	0	26800	
T	0 + 0	70	880	1967 + 2	4	4700	
CA	40 + 0	260	2010	1000 + 137	2	7300	
C	140 + 0	240	1870	397 + 24	12	60700	

<sup>1</sup> The encysted eggs and larvae are not included

TABLE 12. Sandy soil with *H. rostochiensis*. Effect of 5 PSS treatments (H, CP, D, T, CA and C; cf. text – in perspex tubes containing 350 g of soil) on growth of potato and development of the nematodes, evaluated 101 days after planting potato, i.e. on 9.4.1970.

Figures are averages of 10 replicate tubes, weights are fresh weight in g, sizes are in cm, rating indexes are explained in the footnotes. L.S.D. = least significant differences at 5% level; N.S. = no significant difference.

Evaluation results	PSS treatments						L.S.D.
	H	CP	D	T	CA	C	
Total plant weight	2.23	2.98	2.47	2.72	1.81	0.79	0.51
% dry matter	8.1	8.2	8.4	9.3	8.8	8.4	N.S.
Shoot weight	1.52	2.10	1.81	1.86	1.20	1.20	0.55
Shoot size	7.1	8.0	7.2	7.0	5.1	3.1	0.9
Root weight	0.71	0.88	0.66	0.85	0.61	0.24	0.24
Roots as % of total plant weight	31	29	26	31	34	33	N.S.
Index roots size <sup>1</sup>	5.7	7.0	5.9	7.1	5.0	3.2	1.0
Index feeder roots <sup>2</sup>	20.4	20.4	18.8	34.4	22.8	17.2	12.8
Index root colour <sup>3</sup>	6.7	7.3	8.5	5.3	4.7	4.0	0.7
Cysts visible (through plastic <sup>4</sup> )	0.0	0.0	0.0	2.6	15.1	7.0	5.1
Cysts visible on washed roots <sup>5</sup>	0.0	0.0	0.0	10.0	26.0	23.0	7.3
Males and larvae in roots	0	0	0	48	120	85	63
Free larvae in soil (log.)	0 (0.0)	0 (0.0)	0 (0.0)	1971 (3.15)	1137 (2.81)	421 (2.52)	(0.227)
Free males in soil (log.)	0 (0.0)	0 (0.0)	0 (0.0)	2 (0.20)	157 (1.89)	24 (1.20)	(0.33)

<sup>1</sup> – <sup>5</sup> are evaluation figures, estimated jointly by two observers.

<sup>1</sup> Index roots, from 10 (= maximum growth obtained) to 0 (= no roots).

<sup>2</sup> Index feeder roots, from 40 (= maximum density at all the four sides of the square plastic tube) to 0 (= no feeder roots visible).

<sup>3</sup> Index root colour, from 10 (= white) to 0 (= dark brown to black).

<sup>4</sup> Cysts counted through the plastic tube wall.

<sup>5</sup> Cysts counted on the roots after lifting and washing the plants.

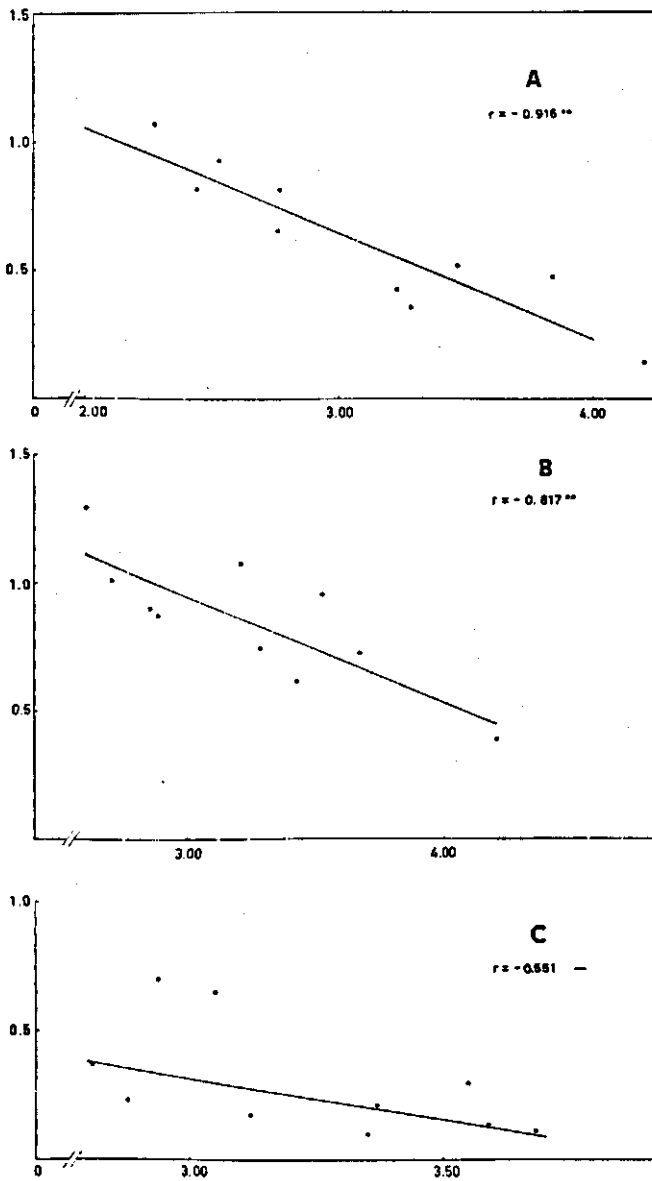


FIG. 10. Regression of root weight of potato on *H. rostochiensis* larvae in the soil 101 days after planting in replicates of three different treatments, viz. Captan (A), Temik (B) and check (C).  $r$  = regression coefficient, — = not significant, \* = significant at 5% level, \*\* = significant at 1% level. Abscissa for each graph: log. number of free larvae in soil, calculated per g of root. Ordinate for each treatment: root weight in g.

$P_1$  in the Table 11 indicates that H, CP and D have eradicated all nematodes. T has affected the population, although a number of 'Other Tylenchida' and 'Saprozoic nematodes' survived. It remains uncertain from this table whether CA has killed or influenced the nematodes.  $P_2$  confirms the eradication of plant nematodes by H, CP and D, but also proves that part of the *H. rostochiensis* population survived the treatments with T and CA and reproduced. The figures for T and CA are even higher than for the untreated C. This may be due to increased reproduction or extended activity on the better developed plants, for the data of Table 12 indicate that partial kill of the *H. rostochiensis* population had originally been reached. It is remarkable that the saprozoic nematodes re-established their populations and reached great densities in the soil treated with H, CP and D as well, but not in the soil treated with T and CA. These differences are marked, but cannot be explained with confidence.

Table 12 confirms the results of Table 11 with respect to the complete kill of *H. rostochiensis* by H, CP and D. No trace of infestation of the plants and no active larvae or males in the soil could be detected, and the better plant growth for CP as compared to H and D of about 15% cannot be related to damage by *H. rostochiensis*. It is noteworthy that the number of cysts and dead eggs in the cysts was still high in the soils treated by H, CP and D even after cultivation of potato. PSS obviously kills and preserves these eggs in the cysts for several months, and they may erroneously be counted as normal eggs in routine laboratory enumerations. This phenomenon was already described by OOSTENBRINK in 1950, who for this reason considered direct counting of eggs in cysts invalid for the evaluation of the effects of nematicides.

The figures for T, CA and C reveal, that the T plants were lighter and that the CA plants were heavier infested than C, i.e. the untreated control plants. This was obvious from the cysts on the outside of the root-surfaces visible through the plastic tube walls, from the cysts on the roots when freed from soil, from the numbers of males and larvae in the roots and from the numbers of free males in the soil, but not from the numbers of free larvae in the soil which were especially

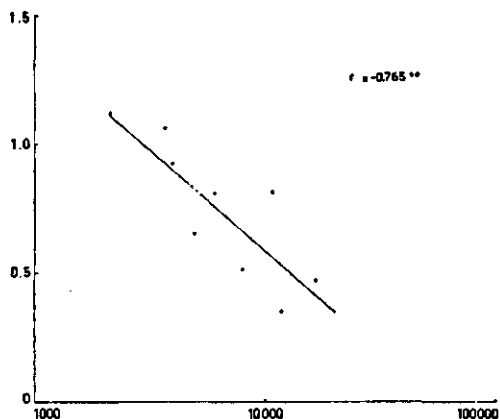


FIG. 11. Regression of root weight of potato on eggs + larvae of *H. rostochiensis* in the soil 101 days after planting within 8 replicates of the soil treated with CA (2 replicates were used for earlier tests).  $r$  = regression coefficient, \*\* = significant at 1% level. Abscissa: log. number of eggs + larvae per tube. Ordinate: root weight in g.

high for T. T apparently has killed or inactivated part of the nematodes (cf. also Table 11), but it has allowed the surviving nematodes to reproduce fairly well as compared to control. The greater final density of free larvae in the T soil may indicate extended activity of the population on the vigorously growing T plants, or may indicate a repellent effect of T. It cannot be indicated whether CA has originally killed some *H. rostochiensis*. It has at any rate increased potato growth and the correspondingly greater food source must be the reason for the strong reproduction on CA plants as compared to control.

The pathogenicity of *H. rostochiensis* can be illustrated within the series of replicates of each treatment, owing to the natural variability between replicates. This is especially so for the treatments from which measurable densities survived viz. CA, T and C. Fig. 10 illustrates the correlations between potato root weight and final larval density in the soil per g of root, for the soils treated with CA, T and C; Fig. 11 shows the regression of potato root weight on total number of eggs and larvae in the soil treated with CA in the same trial. The graphs indicate that *H. rostochiensis* was the dominant factor for potato growth in these soils and, therefore, must have caused much of the average growth differences between the treatments as well.

There are reasons to stress some special results obtained for Temik. Temik did not kill *H. rostochiensis* as efficiently as H or D, but it promoted growth more, about as good as CP. It may therefore be that it protects the plant against nematode infestation, despite the fact that the larvae are present. It may also have other special effects on the plant: the amount of root hairs were significantly higher than for any other treatment, whereas the total root weight and dry matter content of the plant were also higher. On the other hand the roots were less white than for H, CP and D.

### 6.3. CLAY SOIL WITH *H. GOETTINGIANA* UNDER PEA

The soil was a clay soil transported from an infested field at Borssele, Zeeland to an experimental plot at Wageningen in 1957, where it was yearly grown with pea. It comprised a noxious population of *H. goettingiana* and has, in addition, built up a complex of pathogenic fungi (*Phoma medicaginis* var. *pinodella*, *Fusarium* sp. and *Pythium* sp.).

The direct effect of the PSS treatments on the nematodes was partially measured by determining the densities of active nematodes in the soil just before and just after planting, i.e.  $P_i$  and  $P_f$  as described under 6.2. The results are summarized in Table 13. The effects of the treatments on growth of pea and on the *H. goettingiana* infestation are recorded in Table 14 and Fig. 12.

Table 14 and Fig. 12 show that all PSS treatments have increased the plant weight 2–3 fold and the pod weight 4–9 fold. The effects are visible for nearly all statistics in Table 14. The root/total plant ratio of the untreated C is higher, which indicates stagnation of the growth after most roots had been formed, and the dry matter content in the shoots is also relatively high, probably due to

TABLE 13. Clay soil with *H. goettingiana*. Effect of 5 PSS treatments (H, CP, D, T, CA and C; cf. text) on active nematodes in the soil between treatment and planting ( $P_t$ ) and 71 days after cultivation of pea, i.e. on 9.3.1970 ( $P_f$ ). Nematode numbers are per tube containing 350 g of soil, as average of 10 replicates. *H. 1.* + ♂ = *Heterodera* larvae and males, *R.* = *Rotylenchus robustus*, *O.* = other stylet-bearing nematodes, *S.* = saprozoic nematodes.

PSS treatments	Nematodes $P_t$				$P_f$			
	<i>H. 1.</i> + ♂ <sup>1</sup>	<i>R.</i>	<i>O.</i>	<i>S.</i>	<i>H. 1.</i> + ♂ <sup>1</sup>	<i>R.</i>	<i>O.</i>	<i>S.</i>
H	0 + 0	0	0	0	0 + 0	0	0	25600
CP	0 + 0	0	0	0	0 + 0	0	0	20100
D	0 + 0	0	0	0	0 + 0	0	0	24300
T	45 + 0	15	75	3255	536 + 10	24	3	20300
CA	90 + 0	275	165	4845	313 + 262	113	5	42700
C	25 + 0	435	300	7050	105 + 27	165	22	38700

<sup>1</sup> The encysted eggs and larvae are not included.

TABLE 14. Clay soil with *H. goettingiana*. Effect of 5 PSS treatments (H, CP, D, T, CA and C; cf. text - in plastic tubes containing 350 g of soil) on growth of pea and development of the nematodes, evaluated 71 days after sowing pea, i.e. on 9.3.1970. Figures are averages of 10 replicate tubes, weights are fresh weights in g unless indicated otherwise, sizes are in cm, rating indexes are explained in the footnotes. L.S.D. = least significant differences at 5% level; N.S. = no significant difference.

<div>Evalu- ation results</div> <div>PSS treatments</div>	H	CP	D	T	CA	C	L.S.D.
Plant length	22.6	19.1	20.7	18.5	19.8	14.9	2.52
Total plant weight	4.29	3.49	3.79	2.37	3.10	1.42	0.792
Shoot weight without pods	1.92	1.52	1.84	1.22	1.54	0.77	0.363
% dry matter shoots	24.8	19.7	21.3	20.5	19.2	23.5	N.S.
Index shoots <sup>1</sup>	8.1	7.5	7.6	5.8	7.0	3.6	1.11
Index leaves <sup>2</sup>	7.1	7.1	7.0	5.2	5.3	2.9	1.06
Pod weight	1.30	0.99	1.20	0.57	0.63	0.15	0.450
Root weight	1.07	0.99	0.86	0.58	0.93	0.50	0.249
Roots as % of total plant weight	24	29	23	25	30	40	6.3
Index roots <sup>3</sup>	7.7	7.6	5.8	4.8	6.2	3.0	1.32
Index feeder roots <sup>4</sup>	24.3	13.2	4.1	2.7	6.3	3.0	3.21
Index root colour <sup>5</sup>	8.9	8.9	6.5	3.0	6.9	2.7	0.91
Index bacterial nodulation <sup>6</sup>	1.0	2.1	4.3	4.1	0.4	1.1	1.01
Nematode index <sup>7</sup>	0.0	0.0	0.0	1.1	3.6	4.9	0.43

<sup>1-7</sup> are evaluation figures, estimated jointly by two observers.

<sup>1</sup> Index shoots, from 10 (= maximum growth obtained) to 0 (= no growth).

<sup>2</sup> Index leaves, from 10 (= maximum growth and best colour obtained) to 0 (= worst).

<sup>3</sup> Index roots, from 10 (= largest root system) to 0 (= no roots).

<sup>4</sup> Index feeder roots, from 40 (= maximum density at all the four sides of the square plastic tube) to 0 (= no feeder roots visible).

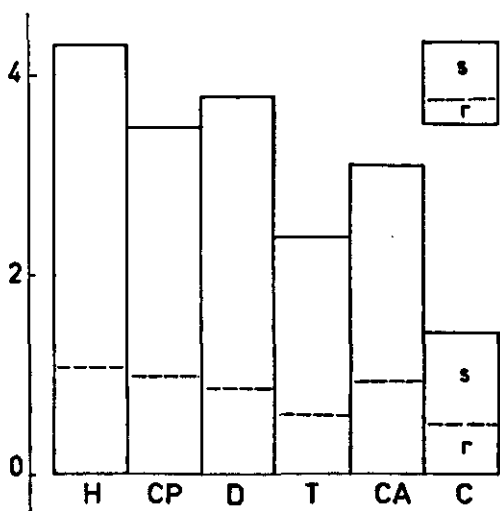
<sup>5</sup> Index root colour, from 10 (= white) to 0 (= dark brown to black).

<sup>6</sup> Index bacterial nodulation, from 5 (= highest degree of nodulation) to 0 (= no nodules).

<sup>7</sup> Nematode index: 0, 1, 2, 3, 4 and 5 indicate 0%, 1-10%, 11-20%, 21-30%, 31-50% and 51-100% of the roots infested with the nematode.



FIG. 12. Effect of different PSS treatments (H, CP, D, T, CA and C; cf. text) on total weight of pea when grown in tubes containing 350 g of soil with *H. goettingiana*. All results are averages of 10 replicate tubes. The least significant differences for s and r are added at the right top corner. Abscissa: PSS treatments as indicated. Ordinate: plant weight in g subdivided in root weight (r) and shoot weight (s).



stagnation of water transport and correspondingly drying up of the plants. The treatments differ in their effects: they decrease in the order H, D, CP, CA, T, C. Difficult to explain are again the influences of T and CA.

Table 13 proves that H, CP and D killed all nematodes. D was somewhat better than CP but somewhat less effective than heat. Heat was superior to CP in this experiment, which was the reverse in the foregoing one. The little difference between D, H and CP suggests, that the effect was mainly nematode control.  $P_1$  in Table 13 for T, CA and C indicates, that T killed more than half and CA about one third of the nematodes (the *Heterodera*-figures are not informative, because they represent only the few free larvae of the encysted population).

The different effects for T and CA, however, (cf. Fig. 12) indicate that CA exerted special growth promoting influence which T did not. This may be the killing off of parasitic fungi. Earlier experiments, however, suggested that T possessed such growth promoting power as well, and the differences between CA and T therefore indicate pea sensitivity to root infecting fungi. As in the earlier experiment with *H. rostochiensis*, it is remarkable that the saprozoic nematodes establish their population and reach high densities in all soils (although the effect of the treatments is still visible in the  $P_1$ , not only for the saprozoic nematodes, but also for *Rotylenchus* spp. and the other stylet-bearing nematodes and for *H. goettingiana* as well. T causes many free larvae in the soil but suppresses the number of males, whereas CA promotes the number of males. These data suggest that T has a repellent effect on the nematodes without killing them and therefore also blocks reproduction.

It is also noteworthy that the bacterial nodulation of pea, which is known to disappear in plants heavily infested by *H. goettingiana* such as the control plants, is restored by D as well as by T, but not by CA, H and CP. H and CP

may have killed the nodulation bacteria together with the nematodes, CA may not have eliminated enough nematodes, or may also have killed the bacteria. There is no reason to believe that the degree of nodulation was the determining factor for growth of the plants, due to nitrogen production, because a heavy dose of nitrogen was applied to all tubes.

#### 6.4. SANDY POLDER SOIL WITH *HEMICYCLIOPHORA CONIDA* UNDER GRASS

The soil used is a fine sea sand from Wieringermeer polder, from a location known to be infested with the sheath nematode *Hemicycliophora conida* (experimental farm Van Bemmelen-Hoeve). The results obtained are recorded in Tables 15, 16 and in Figures 13 and 14.

Table 15 indicates that H, CP and D have eradicated all plant parasitic nematodes,  $P_i$  was zero for *Hem.*, *P.*, *P.r.*, *O.* and *S.*, and the same was true for  $P_f$  except the saprozoic nematodes *S.* which re-established high densities in the course of the experiment. CA and T also killed part of the population. The  $P_i$  figures suggested that the effect for CA was higher than for T for the ectoparasitic *Hemicycliophora conida* but this was the reverse for the endoparasitic *Pratylenchus neglectus*, for its  $P_f$  was markedly suppressed by T in the soil as well as in the roots. This table suggests that T renders the roots of grass toxic to endoparasites and repels them without directly killing them.

The yield increases in Table 16 and Fig. 13A are marked. They are well in agreement with the elimination of *Hem. conida*, except for D which probably had some toxic effect on the plants. Fig. 13B confirms the result illustrated earlier in Fig. 8, namely that the differences in plant growth due to treatments usually increase with time. Fig. 14 illustrates a highly significant regression of root weight of grass on the final *Hem. conida* density in the soil per gram roots. This suggests that the nematode was the main cause of the differences in root growth; the evidence is strong considering the fact from other experiments that the nematode is known to be noxious to grass.

TABLE 15. Sandy polder soil with *Hemicycliophora conida*. Effect of 5 PSS treatments (H, CP, D, T, CA and C; cf. text) on active nematodes in the soil and in the roots between treatment and planting ( $P_i$ ) and 90 days after sowing ryegrass, i.e. on 30.3.1970 ( $P_f$ ). Nematode numbers are per tube containing 350 g of soil, as average of 10 replicates. *Hem.* = *Hemicycliophora conida*, *P.* = *Pratylenchus neglectus*, *P.r.* = *P. neglectus* inside the roots, *O.* = other stylet-bearing nematodes, *S.* = saprozoic nematodes.

PSS treatments	Nematodes				$P_i$					$P_f$				
	<i>Hem.</i>	<i>P.</i>	<i>O.</i>	<i>S.</i>	<i>Hem.</i>	<i>P.</i>	<i>P.r.</i>	<i>O.</i>	<i>S.</i>	<i>Hem.</i>	<i>P.</i>	<i>P.r.</i>	<i>O.</i>	<i>S.</i>
H	0	0	0	0	0	0	0	0	0	0	0	0	0	21414
CP	0	0	0	0	0	0	0	0	0	0	0	0	0	29324
D	0	0	0	0	0	0	0	0	0	0	0	0	0	11920
T	1140	1160	330	1450	948	16	4	17	21550					
CA	640	1870	150	2990	77	75	338	342	32000					
C	2000	2080	350	4410	3823	41	123	38	18500					

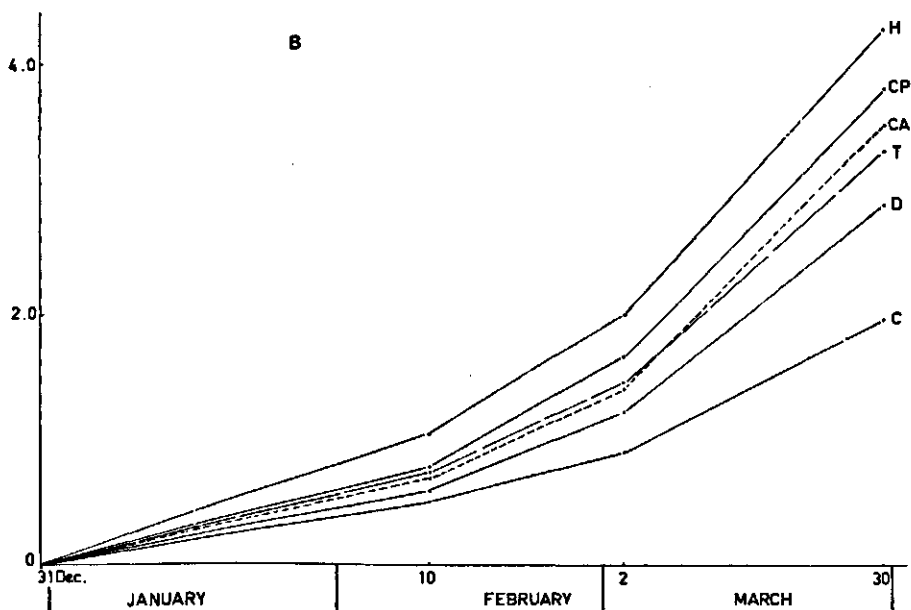
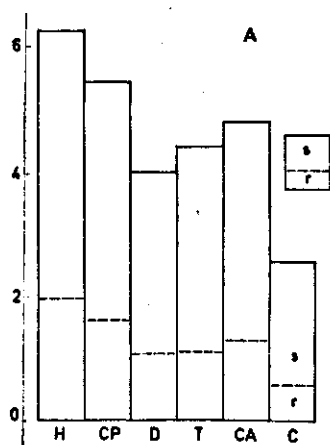


FIG. 13. Effect of different soil treatments (H, CP, D, T, CA and C; cf. text) on grass growth in tubes containing 350 g of soil with *Hem. conida*. All results are averages of 10 replicate tubes. A. Abscissa: PSS treatments as indicated. The least significant differences for *s* and *r* are added at the right top corner. Ordinate: plant weight subdivided in root weight (*r*) and shoot weight (*s*). B. Abscissa: time scale, mentioning sowing date and three cutting dates. Ordinate: grass yield in g per tube, cumulative.

TABLE 16. Sandy polder soil with *Hem. conida*. Effect of 5 PSS treatments (H, CP, D, T, CA and C; cf. text) in plastic tubes containing 350 g of soil on growth of ryegrass and development of the nematodes, evaluated 90 days after sowing the grass, i.e. on 30.3.1970. Figures are averages of 10 replicate tubes; weights are fresh weight in g, rating indexes are explained in the footnotes 3, 4 and 5 of Table 14. L.S.D. = least significant differences at 5% level; N.S. = no significant difference.

Evaluation results	PSS treatments	H	CP	D	T	CA	C	L.S.D.
Total plant weight		6.24	5.43	3.98	4.41	4.80	2.54	0.575
Total shoot weight, cuts I + II + III		4.27	3.81	2.89	3.30	3.51	1.95	0.425
% dry matter of total shoot weight		15.0	14.6	14.6	14.6	14.4	14.4	N.S.
Root weight		1.97	1.62	1.09	1.12	1.29	0.59	0.227
Roots as % of total plant weight		31.4	29.4	27.4	24.9	26.3	21.5	3.30
Index roots <sup>1</sup>		9.0	7.4	5.2	5.8	6.0	2.7	0.76
Index feeder roots <sup>2</sup>		24.4	13.0	12.9	12.7	13.5	7.2	3.20
Index root colour <sup>3</sup>		6.0	6.1	8.0	6.2	6.9	6.4	N.S.

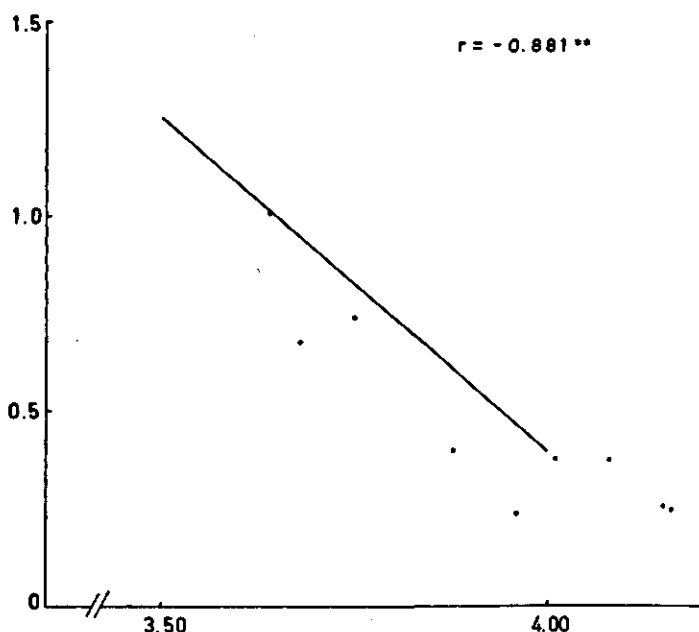


FIG. 14. Regression of root weight of ryegrass on final *Hem. conida* density in the soil 90 days after sowing within replicates of the untreated control C.  $r$  = regression coefficient; \*\* = significant at 1% level. Abscissa: log. number of *Hem. conida* in the soil, calculated per g of roots. Ordinate: root weight in g.

## 6.5. SANDY GLASSHOUSE SOIL WITH *MELOIDOGYNE JAVANICA* UNDER TOMATO

The soil was a sandy soil kept in the PD glasshouse. It was infested with a culture of *M. javanica* and several crops of tomato were then grown in succession. The PSS effect on the nematode population was strong, according to  $P_i$  and  $P_f$  of the active nematode stages in Table 17. It should, however, be kept in mind that extraction of active nematodes does not measure the *Meloidogyne* potential in root debris. Growth of the tomato test plant did not react correspondingly to the nematodes; it was increased measurably by H, CP and CA, but not by D and T, according to Table 18 and Fig. 15.

Table 17 illustrated near-complete nematicidal effects of H, CP and D. T obviously only had a slight direct effect, according to  $P_i$ , but the low value of  $P_f$  indicates that the treatment finally reduced the *Meloidogyne* spp. densities in soil and in roots nearly as much as H, CP and D. This indicates again a prolonged, perhaps a systemic nematicidal effect of T. The extremely low density of the saprozoic nematodes, only for T, accentuates its special nematicidal influence. CA was apparently ineffective in reducing the nematode population.  $P_f$  of *Meloidogyne* in soil and in roots was even considerably higher than for untreated C, probably owing to better development of the plants and absence of competing fungi in the soil.

Fig. 15 and Table 18 show that the growth promoting effects of the treatments were probably not due to elimination of *M. javanica*, the only plant nematode numerously present in this soil, since D and also T suppressed the population effectively but did not cause growth increase, as H and CP did. Furthermore CA did not suppress the nematodes at all, but promoted plant growth nearly as much as H and CP. The effects of H, CP and CA are possibly due to suppression of parasitic fungi, which may have been of more significance than *M.*

TABLE 17. Sandy glasshouse soil with *M. javanica*. Effect of 5 PSS treatments (H, CP, D, T, CA and C, cf. text) on active nematodes in the soil between treatment and planting ( $P_i$ ) and 92 days after cultivation of tomato, i.e. on 6.5.1970 ( $P_f$ ). Nematode numbers are per tube of 350 g of soil as averages of 9 replicates. *M. 1.* = *Meloidogyne* larvae, between brackets the corresponding numbers of *M. 1.* extracted from the roots by incubation, *O.* = other stylet-bearing nematodes, *S.* = saprozoic nematodes.

PSS treatments	nematodes			$P_i$			$P_f$		
	<i>M.1.</i>	<i>O.</i>	<i>S.</i>	<i>M.1.</i>	<i>O.</i>	<i>S.</i>	<i>M.1.</i>	<i>O.</i>	<i>S.</i>
H	0	0	1	0	(0)	0	12300		
CP	0	0	2	0	(0)	0	15900		
D	0	0	1	0	(3)	0	18600		
T	230	10	21000	26	(21)	0	410		
CA	860	100	26000	6309	(6363)	0	9800		
C	750	140	18000	1439	(1110)	0	9000		

TABLE 18. Sandy glasshouse soil with *M. javanica*. Effect of PSS treatments (H, CP, D, T, CA and C; cf. text) in plastic tubes with 350 g of soil on the growth of tomato and the development of the nematodes, evaluated 92 days after planting, i.e. 6.5.1970. Figures are averages of 9 replicate tubes, weights are fresh weight in g, rating indexes are explained in the footnotes 5 and 7 of Table 14. L.S.D. = least significant differences at 5% level; N.S. = no significant difference.

Evaluation results	PSS treatments						L.S.D.
	H	CP	D	T	CA	C	
Total plant weight	5.77	6.11	4.53	4.57	5.80	4.34	1.23
Shoot weight	3.55	3.86	2.92	2.96	3.74	2.75	0.82
% dry matter of shoot weight	13.8	14.0	14.0	15.9	15.1	16.3	N.S.
Root weight	2.22	2.25	1.61	1.61	2.06	1.59	0.56
% root weight of total plant weight	36.4	36.3	33.9	33.8	34.3	36.2	N.S.
Root colour index <sup>1</sup>	6.6	7.1	7.1	7.2	5.6	5.3	0.8
Root gall index <sup>2</sup>	0.0	0.0	0.0	1.1	5.0	3.8	0.4

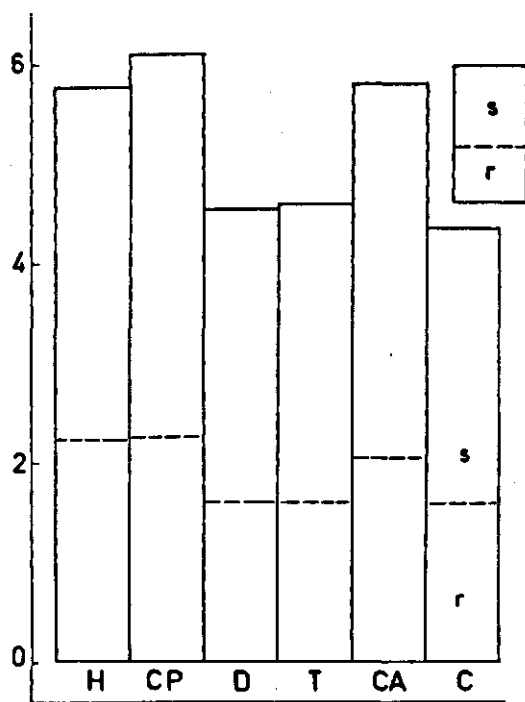


FIG. 15. Effect of different PSS treatments (H, CP, D, T, CA and C; cf. text) on total weight of tomato plants when grown in tubes containing 350 g of soil with *M. javanica*. All results are averages of 9 replicate tubes. The least significant differences for r and s are added at the right top corner. Abscissa: PSS treatments as indicated. Ordinate: plant weight in g subdivided in root weight (r) and shoot weight (s).

*javanica* in this soil. The fact, however, that the roots of the plants for D and T had a healthy white colour, whereas those for CA were nearly as brown as for C weakens this hypothesis. Further, the author had previously observed, that CA plants, despite vigorous growth, did not give a satisfactory fruit yield like those with H and CP treatments. Therefore with certainty elimination of noxious root fungi cannot be indicated as the cause of growth responses observed. The possibility that D and T had been slightly toxic and that CA had initially delayed nematode infestation or compensated it by stimulating plant growth cannot be excluded.

The conclusion is therefore that H, CP and CA obtained moderate growth responses in contrast to D and T. The cause of these responses is obviously not related to *M. javanica* in this stage of plant growth, and specific tomato-root infecting fungi which were not eliminated by D, T and C seemed to have played a significant role.

#### 6.6. POLDER CLAY SOIL WITH *MELOIDOGYNE NAASI* UNDER CABBAGE

The soil was a sample from a clay area in Oostelijk Flevoland where *M. naasi* was present.

The PSS effect on the nematode population was great in some and negligible in other treatments and is summarized in Table 19. Growth of the test plant cabbage was greatly increased by some but even depressed by other treatments, according to Table 20 and Fig. 16.

Table 19 shows approximately the same effects as described in the experiment under 6.5. H, CP and D caused almost complete eradication of the nematodes. T may initially have had some effect, considering  $P_i$  for *O.* and *S.* The fact that  $P_i$  for *M.1.* is higher may be due to stimulation and hatch of larvae from the population in root debris and need not be a determining factor in this case. The low value of  $P_f$  for *M.1.*, in agreement with the relatively low  $P_f$  value for *O.* and *S.* as compared to C, confirms this impression. CA was per-

TABLE 19. Polder clay soil with *M. naasi*. Effect of 5 PSS treatments (H, CP, D, T, CA and C; cf. text) on active nematodes in the soil between treatments and planting ( $P_i$ ) and 72 days after cultivation of cabbage, i.e. on 11.5.1970 ( $P_f$ ). Nematode numbers are per tube containing 350 g of soil as average of 11 replicates. *M.1.* = free *Meloidogyne* larvae, *O.* = other stylet-bearing nematodes and *S.* = saprozoic nematodes.

PSS treatments	Nematodes					
	$P_i$			$P_f$		
	<i>M.1.</i>	<i>O.</i>	<i>S.</i>	<i>M.1.</i>	<i>O.</i>	<i>S.</i>
H	0	0	5	0	0	12000
CP	0	0	0	0	0	13200
D	0	0	1	0	0	4900
T	200	230	1340	1	97	7100
CA	80	215	2000	2	242	22000
C	20	386	6447	33	134	9600

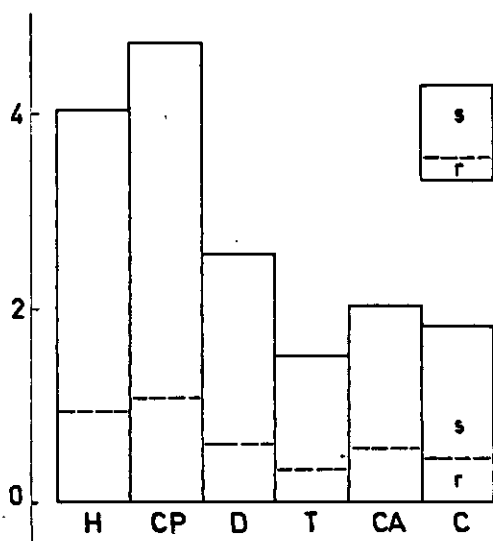


FIG. 16. Effect of different PSS treatments (H, CP, D, T, CA and C; cf. text) on total weight of cabbage plants when grown in tubes containing 350 g of soil with *M. naasi*. All results are averages of 11 replicate tubes. The least significant differences for r and s are added at the right top corner. Abscissa: PSS treatments as indicated. Ordinate: plant weight in g subdivided in root weight (r) and shoot weight (s).

TABLE 20. Clay soil with *M. naasi*. Effect of 5 PSS treatments (H, CP, D, T, CA and C; cf. text) in plastic tubes containing 350 g of soil on growth of cabbage, evaluated 72 days after sowing, i.e. on 11.5.1970. Figures are averages of 11 replicate tubes, weights are fresh weight in g unless indicated otherwise, indexes are explained in the footnotes. L.S.D. = least significant differences at 5% level.

Evaluation results	PSS treatments	H	CP	D	T	CA	C	L.S.D.
Total plant weight		4.02	4.69	2.53	1.49	2.00	1.79	0.96
% dry matter of plant weight		17.6	17.7	15.4	13.5	17.0	13.8	1.71
Shoot weight		3.09	3.60	1.95	1.16	1.45	1.35	0.76
Index flowers <sup>1</sup>		1.0	1.0	0.3	0.0	0.4	0.0	0.23
Root weight		0.93	1.09	0.59	0.33	0.55	0.44	0.22
Index root colour <sup>2</sup>		6.5	5.9	6.9	7.1	5.6	6.6	0.80

<sup>1</sup>Index flowers, from 1 (= all tubes with flowers) to 0 (= no tubes with flowers).

<sup>2</sup>Index root colour, from 10 (= white) to 0 (= dark brown to black).

haps initially somewhat effective, but the final populations were, as in all earlier experiments, again higher than for C. The successful recovery of the saprozoic nematodes for H, CP and D, as well as the very high value for CA, are noteworthy.

Table 20 and Fig. 16 illustrate some unexpected results. Plant growth was strongly promoted by H and CP, moderately by D, hardly by CA and was depressed by T. T has obviously caused phytotoxicity (combined with a low dry matter content), and this may be the same for D. Root colour was optimum for T and D and minimum for CA. The fact that phytotoxicity played a role,



and that CA had no effect, makes it probable that the growth promotions are largely due to the elimination of nematodes and not fungi. It is possible that, in addition to *Meloidogyne naasi*, the other stylet-bearing nematodes, mainly *Paratylenchus* sp. and *Criconebella parva*, have contributed to the growth differences observed.

It is obvious that cabbage suffered a marked growth deficiency in this soil, and it is probable that nematode damage was involved. The results, however, do not give a clear conclusion, the more so because phytotoxicity was at any rate involved.

#### 6.7. POLDER CLAY SOIL WITH *MELOIDOGYNE NAASI* UNDER BARLEY

Another polder soil from Oostelijk Flevoland infested with *M. naasi* was used for a test with barley. Fig. 17 and Tables 21 and 22 represent the effect of the PSS treatments on the density of active nematodes in the soil and the plant reaction.

H, CP and D have suppressed the nematodes effectively, so that  $P_i$  as well as  $P_f$  of the stylet-bearing nematodes was zero (Table 21). The saprozoic forms, however, re-established a moderate population level. T and CA reduced the final root-knot nematode larval density as compared to C; they may have suppressed the initial population. T has at any rate been more effective than CA.

The plant response to T and CA was negative, which must be due to phytotoxic effects of the treatments (Table 22, Fig. 17).

CP promoted growth more than H and D, according to most of the criteria used; H was usually intermediate between CP and D. The differences between CP, H and D cannot be related to nematode infestation. The gall index confirmed the strong nematicidal effects of H, CP and D and also that T had been more effective than CA, and CA more effective than C, as already indicated

TABLE 21. Polder clay with *M. naasi*. Effect of 5 PSS treatments (H, CP, D, T, CA and C; cf. text) on active nematodes in the soil between treatment and planting ( $P_i$ ) and 90 days after cultivation of barley, i.e. on 29.3.1970 ( $P_f$ ). Nematode numbers are per tube containing 350 g of soil as averages of 9 replicates. M.I. = free *Meloidogyne* larvae, between brackets the corresponding numbers of M.I. extracted from the roots by incubation, O. = other stylet-bearing nematodes, S. = saprozoic nematodes.

PSS treatments	Nematodes					
	$P_i$			$P_f$		
	M.I.	O.	S.	M.I.	O.	S.
H	0	0	0	0 (0)	0	5800
CP	0	0	0	0 (0)	0	10000
D	0	0	0	0 (0)	0	1500
T	318	405	1200	24 (4)	53	7100
CA	1930	3400	7810	117 (37)	167	36200
C	1740	4500	8600	430 (104)	1406	93400

TABLE 22. Polder clay soil with *Meloidogyne naasi*. Effect of PSS treatments (H, CP, D, T, CA and C; cf. text) in plastic tubes containing 350 g of soil on growth of barley and development of the nematodes, evaluated 90 days after sowing the barley, i.e. on 29.3.1970. Figures are averages of 9 replicate tubes, weights are fresh weight in g, sizes are in cm, rating indexes 1-4 are explained in the footnotes 3, 4, 5 and 7 of Table 14. L.S.D. = least significant differences at 5% level.

Evaluation results \ PSS treatments	H	CP	D	T	CA	C	L.S.D.
Plant length	64	69	64	54	44	57	5.9
Total plant weight	9.9	12.2	8.5	4.5	3.7	6.6	1.58
Shoot weight	7.9	10.2	6.9	3.7	3.1	5.3	1.37
Number of flowers	1.8	1.6	1.2	0.0	0.0	0.2	0.51
Root weight	1.98	2.05	1.60	0.85	0.62	1.35	0.36
Index root size <sup>1</sup>	7.0	6.4	5.5	3.3	2.2	5.7	0.81
Index feeder roots <sup>2</sup>	15.8	13.9	12.0	6.2	4.0	11.9	3.82
Index root colour <sup>3</sup>	5.7	5.0	6.0	2.9	1.8	4.3	0.79
Root gall index <sup>4</sup>	0.0	0.0	0.0	1.1	2.6	3.8	0.94

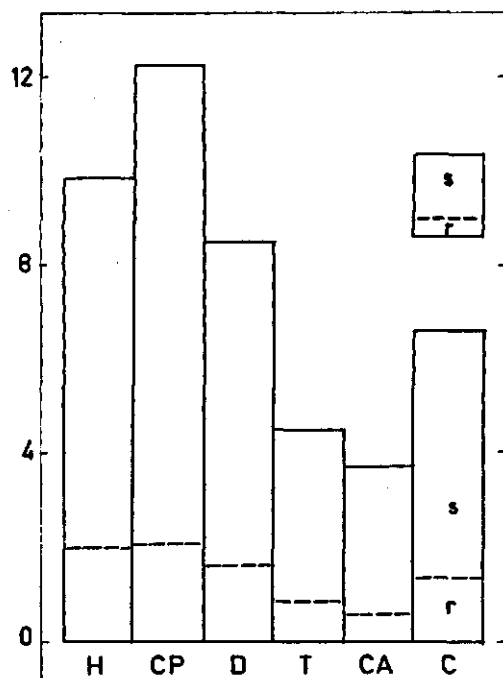


FIG. 17. Effect of different PSS treatments (H, CP, D, T, CA and C, cf. text) on total weight of barley plants when grown in tubes containing 350 g of clay soil with *M. naasi*. All results are averages of 9 replicate tubes. The least significant differences are added at the right top corner. Abscissa: PSS treatments as indicated. Ordinate: plant weight in g subdivided in root weight (r) and shoot weight (s).

by the data of Table 21. The gall index confirmed that the poor growth of T and CA compared to C was not due to root-knot infestation.

It is, therefore, probable that most of the plant growth differences between C and the three successful treatments H, CP and D were due to the elimination of nematodes, probably primarily *M. naasi*, but the other marked differences (CP versus H and D, T and CA versus C) were not.

## 6.8. SANDY POLDER SOIL WITH *MELOIDOGYNE NAASI* UNDER BARLEY

A sandy polder soil from Middenmeer, Oostelijk Flevoland, with a marked infestation of *M. naasi*, was used for another test with barley. Fig. 18 and Tables 23 and 24 show the results.

The eradication of the active nematodes by H, CP and D was as complete as in the experiment under 6.7.  $P_f$  suggests that T and CA also had suppressed the nematode population. This may be due, as in the experiment under 6.7. to the relatively poor development of the test plant.

Plant growth figures indicate that T and CA have been toxic to barley. H, CP and D increased plant growth to about the same high degree, although CP was slightly superior to the others. This is true for nearly all plant statistics recorded in Table 24. The lower plant response to H treatment comparable to CP seems to be due to unbalanced minerals in the soil. Thermal treatment as an artificial weathering factor does liberate some elements in the soil which may not be desired by some plants. For this reason the differences between H and CP obtained are greater in the rich clay (6.7.) than in the poor sandy soil.

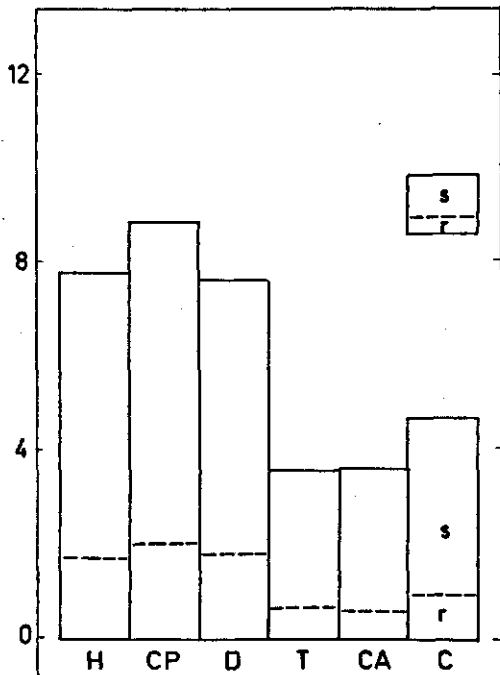


FIG. 18. Effect of different PSS treatments (H, CP, D, T, CA and C, cf. text) on total weight of barley plants when grown in tubes containing 350 g of sandy soil with *M. naasi*. All results are averages of 9 replicate tubes. The least significant differences for r and s are added at the right top corner. Abscissa: PSS treatments as indicated. Ordinate: plant weight in g subdivided in root weight (r) and shoot weight (s).

TABLE 23. Sandy polder soil with *M. naasi*. Effect of 5 PSS treatments (H, CP, D, T, CA and C, cf. text) on active nematodes in the soil between treatment and planting ( $P_i$ ) and 90 days after cultivation of barley, i.e. on 29.3.1970 ( $P_f$ ). Nematode numbers are per tube containing 350 g of soil as average of 9 replicates. *M.1.* = free *Meloidogyne* larvae, between brackets the corresponding numbers of *M.1.* extracted from the roots by incubation, *O.* = other stylet-bearing nematodes, *S.* = saprozoic nematodes.

PSS treatments	Nematodes			$P_i$			$P_f$		
	<i>M.1.</i>	<i>O.</i>	<i>S.</i>	<i>M.1.</i>	<i>O.</i>	<i>S.</i>	<i>M.1.</i>	<i>O.</i>	<i>S.</i>
H	0	0	0	0 (0)	0	14100	0 (0)	0	9300
CP	0	0	0	0 (0)	0	6400	0 (0)	0	3500
D	0	0	0	0 (0)	0	9700	0 (0)	0	24400
T	200	180	2280	26 (2)	0	3500	12 (3)	0	9700
CA	240	220	1570	12 (3)	0	9700	86 (3)	10	24400
C	210	60	3255	86 (3)	10	24400			

TABLE 24. Sandy polder soil with *Meloidogyne naasi*. Effect of PSS treatments (H, CP, D, T, CA and C; cf. text) in plastic tubes containing 350 g of soil on growth of barley and development of the nematodes, evaluated 90 days after sowing the barley, i.e. on 29.3.1970. Figures are averages of 9 replicate tubes, weights are fresh weight in g, sizes are in cm, rating indexes 1-4 are explained in the footnotes 3, 4, 5 and 7 of Table 14. L.S.D. = least significant differences at 5% level.

	PSS treatments						
	H	CP	D	T	CA	C	L.S.D.
Evaluation results							
Plant length	61.3	64.7	59.6	52.6	51.1	55.2	8.47
Total plant weight	7.82	8.88	7.66	3.62	3.64	4.75	1.16
Shoot weight	6.05	6.84	5.80	2.91	3.03	3.79	0.91
Number of plants	4.1	4.5	3.8	2.9	3.8	3.9	0.48
Number of flowers	1.0	1.2	0.8	0.0	0.3	0.0	0.48
Root weight	1.77	2.04	1.86	0.71	0.61	0.96	0.346
Index root size <sup>1</sup>	6.7	7.3	6.7	3.0	2.5	3.6	0.86
Index feeder roots <sup>2</sup>	12.4	11.7	11.0	6.6	1.6	8.8	4.31
Index root colour <sup>3</sup>	7.3	8.2	8.6	4.0	3.0	3.9	0.89
Root gall index <sup>4</sup>	0.0	0.0	0.0	1.0	1.0	1.4	0.43

#### 6.9. LOAMY SAND (PD) SOIL WITH A COMPLEX OF *PRATYLENCHUS CRENATUS* + *TYLENCHORHYNCHUS DUBIUS* + *ROTYLENCHUS ROBUSTUS* UNDER OAT

Soil from the PD garden with the fore-mentioned complex was treated, and grown with oat as a test plant. Fig. 19 and Tables 25 and 26 represent the results.

H, CP and D were again the best treatments for nematode control as in the earlier experiments. T also resulted in a low *P. crenatus* population in the soil as well as in the root tissues and lowered the density of saprozoic nematodes

specifically as well. CA resulted in lower densities for all plant nematodes in the soil, but the *Pratylenchus* density in the roots was as high as for the control plants.

All treatments increased plant growth significantly, but H was far superior to the others. It is remarkable that CA was as effective as CP, D and T. The damage may comprise a nematode and a fungus factor and CA may have specially affected the fungi: the fact that roots of CA plants had a better colour than for any other treatment supports this supposition. It would then, however, be normal if CP had been superior to D, which was not the case due to its plants prematurity as % dry matter indicates.

The result must therefore be complex and cannot fully be explained on the basis of this experiment, cf. also Chapter 8.

TABLE 25. PD sandy soil with a complex of *Pratylenchus crenatus* + *Tylenchorhynchus dubius* + *Rotylenchus robustus*. Effect of PSS treatments (H, CP, D, T, CA and C; cf. text) on active nematodes in the soil between treatment and planting ( $P_i$ ) and 63 days after cultivation of oat, i.e. on 23.4.1970 ( $P_f$ ). Nematode numbers are per tube containing 350 g of soil as average of 10 replicates.  $P$ . = *P. crenatus*, between brackets the corresponding numbers of *P* extracted from the roots,  $T$ . = *T. dubius*,  $R$ . = *R. robustus*,  $O. + S.$  = other stylet bearing nematodes + saprozoic nematodes.

Treatments	Nematodes							
	$P_i$				$P_f$			
	$P$ .	$T$ .	$R$ .	$O. + S.$	$P$ .	$T$ .	$R$ .	$O. + S.$
H	0	0	0	0	0	(0)	0	0 18400
CP	0	0	0	2	0	(0)	0	0 10900
D	0	0	0	3	0	(0)	0	0 14100
T	117	395	178	1371	41	(14)	36	160 1950
CA	293	248	358	810	121	(252)	45	134 24400
C	430	585	570	2280	634	(222)	1211	361 16950

FIG. 19. Effect of PSS treatments (H, CP, D, T, CA and C; cf. text) on total weight of oat when grown in tubes containing 350 g of soil. All results are averages of 10 replicate tubes. The least significant difference for  $r$  and  $s$  are added at the right top corner. Abscissa: PSS treatments as indicated. Ordinate: plant weight in g subdivided in root weight ( $r$ ) and shoot weight ( $s$ ).

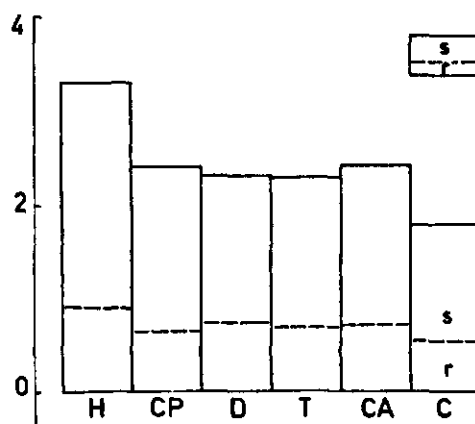


TABLE 26. Loamy sand soil with a complex of *Pratylenchus crenatus* + *Tylenchorhynchus dubius* + *Rotylenchus robustus*. Effect of PSS treatments (H, CP, D, T, CA and C; cf. text) in plastic tubes containing 350 g of soil on the growth of oat and development of the nematodes, evaluated 63 days after sowing the oats, i.e. on 23.4.1970. Figures are averages of 10 replicate tubes; weights are fresh weight in g, sizes are in cm, rating indexes 1-2 are explained in the footnotes 3 and 5 of Table 14. L.S.D. = least significant differences at 5% level.

Evaluation results	PSS treatments						L.S.D.
	H	CP	D	T	CA	C	
Plant length	35	39	30	32	41	27	4.5
% dry matter shoots	19.4	17.8	20.3	17.8	16.1	20.6	1.99
Total plant weight	3.3	2.4	2.3	2.3	2.4	1.8	0.40
Shoot weight	2.4	1.7	1.6	1.6	1.7	1.3	0.28
Root weight	0.92	0.66	0.74	0.69	0.71	0.52	0.14
Index root size <sup>1</sup>	6.8	6.2	6.3	5.5	5.0	3.2	0.69
Index root colour <sup>2</sup>	7.4	6.7	6.9	6.8	8.1	3.9	0.37
<i>P.</i> in roots (log)	0.0	0.0	0.0	0.82	2.29	2.21	0.28
<i>P.</i> in soil (log)	0.0	0.0	0.0	1.53	1.81	2.68	0.31
<i>T.</i> in soil (log)	0.0	0.0	0.0	1.49	1.48	2.96	0.21
<i>R.</i> in soil (log)	0.0	0.0	0.0	2.06	2.06	2.50	0.21

## 6.10. DISCUSSION

The application of selected PSS treatments to nematode-infested soils has led to yield increase results according to expectations, except for two soils in which nematodes were not the main (cf. Fig. 15) nor the only problem for the test plant (cf. Fig. 19), also except for the phytotoxic effects in some of the soils by T and CA (cf. Figs. 16, 17, 18) as well as by D (cf. Figs. 13, 16). In most soils control plants were very poor and nematicidal treatments were effective in removing the plant growth deficiency cause.

Growth differences between treatments were often great. They could usually be measured by total weight, size or other evaluation statistics listed in the tables. The growth deficiency removed by the treatments was therefore a general delay in the growth of the plants, resulting in reduction of plant length, weight of shoots, roots and fruits, number of roots and flowers, and other quantitative characters, but seldom resulting in a change of the relative characters. Incidentally, however, also the percentage of dry matter (Table 20) or the percentage of roots (Table 16) were influenced.

H and CP eradicated the plant nematodes in all soils and caused more growth increase than the other treatments. Both are known to kill all organisms, but CP was nevertheless better than H in some soils (Figs. 9, 16, 17) and less good in others (Figs. 12, 13, 19). These differences must be due to secondary

(probably phytotoxic) effects, which apparently are not negligible for CP (cf. also Chapter 5) and even for H.

Deradicated the plant nematodes as completely as H and CP, and caused as much yield increase as H or CP, except for two soils in which it apparently caused marked phytotoxicity (cf. Figs 13 and 16) and for one soil in which a fungus problem was most probably present (cf. Fig. 15). The slightly lower effect for D as compared to H and CP in the soils with nematode problems may be fully due to occasional phytotoxic effects, its specificity to control only nematodes while other pathogenic micro-organisms may not be influenced, or the sensitivity of some plants to Cl-ions.

T had a less drastic effect on  $P_1$  of the nematodes. Its effect appeared gradually and was extended. It suppressed the number of nematodes in the roots despite high numbers around the roots (Tables 12, 13, 15, 17, 18, 21, 22, 25, 26). These data support the general concept that T primarily acts as a systemic pesticide. Its influence on plant growth was often comparable to the effect by D, but it was considerably less in Fig. 12, and even more so than the untreated C in Figs 16, 17, 18. T must have been phytotoxic in several soils; the differences between the effects between D and T on plant growth may be due to variation in phytotoxicity rather than variation in nematicidal or other disease controlling effects.

CA may have killed part of the nematodes in several of the experimental soils (Tables 15, 21), but its effect must have been less than for T and the other pesticides.  $P_1$  is often higher for CA than for C, probably due to stronger reproduction on better growing test plants (Tables 17, 18) or minimized population of some nematode natural enemies, i.e. predators and nematophagous fungi. Growth of the test plants was promoted about as much as in the other treatments according to Figs 13, 15 and 19; it was less effective according to Figs 9, 12 and 16, and it was strongly phytotoxic according to Figs 17 and 18. CA was among the best treatments only, however, in the soil represented in Fig. 19; D and T were not effective here, probably because the main damage was caused by fungi and not by nematodes. The results confirm that CA can effectively increase plant growth in some soils, presumably soils with noxious fungi. It may also have some nematicidal effect, but this is far less than for T, D, CP, and H.

The significant regression within the replicates of the untreated control or a certain treatment of root weights on  $P_1$  of certain nematodes (Figs 11, 12 and 14) indicates that in these cases the nematodes must have caused much of the yield differences between control and treated soils. There is little doubt, also on the basis of general knowledge, about the importance of nematodes as noxious parasites in all the test soils, except for the soils represented in Fig. 15 (where fungus damage apparently dominated) and in Fig. 19 (where at any rate

a fungus factor may have been involved). The problem in the last mentioned soil will be studied especially in Chapter 8.



## 7. PSS TREATMENTS OF FRESH OR UNINFESTED SOILS OF DIFFERENT TYPES

### 7.1. INTRODUCTION

The effect of PSS treatments appeared to be great on plant growth in regularly cultivated soils (Chapters 4 and 5), especially so when important plant pests were present (Chapter 6). In this chapter soils of widely different types with or without very few pathogenic organisms were chosen, in addition to PD soil for comparison. They were treated in various ways and grown with a number of test plants in large pots to measure the effects on plant growth.

The 4 main soils used in this series of experiments were:

- a. S = pure river sand, taken from the shore of the river Rhine and washed free from soluble elements. The soil had a high pH and it was practically sterile.
- b. F = Flevoland soil. This is a soil mainly consisting of fine marine sand which had not yet been grown to crops and only lightly covered with spontaneous plant growths since its reclamation from the sea 7 years ago. It comprised a fair number of saprozoic nematodes and a few other stylet-bearing nematodes, which were probably fungus feeders, but no known or suspected plant parasitic nematodes.
- c. M = meadow soil mainly consisting of sand and humus. It was taken from an old meadow near Wageningen, exposed to drought and therefore partially sterilized by physical means. The pH was very low. The parasitic nematode population was therefore unnaturally low for a meadow soil, although saprozoic nematodes were still, or again, rather numerous.
- d. PD = PD soil, taken from the garden of the Plantenziektenkundige Dienst. The pH was moderately low. It is a sandy loam with high numbers of *Pratylenchus crenatus*, *Tylenchorhynchus dubius*, *Rotylenchus robustus* (complex nr. 5 in Tables 5, 6, 7), in addition to small numbers of other plant parasites and a moderate density of saprozoic nematodes.

Table 27 comprises figures about the mechanical composition and some other soil properties and about the nematode densities of the layer 0–20 cm, before the treatments were applied and the test plants were grown.

The treatments were as follows:

- a. H = heat, 60°C for two hours as described in 6.1.;
- b. CP = chloropicrin, at a dose comparable to 40 ml/m<sup>2</sup>, injected into the pots with 2½ l soil at one central spot near the bottom. The bottom hole was temporarily closed and the pot was temporarily covered with polythene;
- c. D1 = dichloropropene high dose, in the formulation of DD at 128 ml/m<sup>2</sup>, injected as for CP but sealed by occasionally spraying the top layer with water;
- d. D2 = dichloropropene light dose, viz. DD at 32 ml/m<sup>2</sup>, injected and covered with polythene as for CP;

TABLE 27. Properties of the four main soils used for the experiments under 7.

Soils Characters	S = pure river sand	F = Flevoland soil	M = meadow soil	PD = PD sandy loam
<b>A. Soil properties</b>				
Sand, >16 $\mu$ (between brackets >150 $\mu$ )	96 (91)%	71 (28)%	59 (33)%	88 (22)%
Clay, <16 $\mu$	2.0%	20.0%	8.0%	9.0%
Humus	0.3%	2.5%	33.2%	2.4%
CaCO <sub>3</sub>	1.9%	6.7%	—	—
pH-Kc.l	7.6	7.5	4.3	5.0
<b>B. Active nematodes per 100 ml of soil, present before PSS<sup>1</sup></b>				
<i>Pratylenchus</i>	0	0	0	1221
<i>Tylenchorhynchus</i>	0	0	4	893
<i>Rotylenchus</i>	0	0	7	1001
<i>Heterodera</i> larvae	0	0	1	0
Other stylet-bearing nematodes	0	15	200	237
Saprophytic nematodes	15	970	1295	1144

<sup>1</sup> The infective potential of *Heterodera* eggs and larvae in cysts was not determined.

e. PCNB = pentachloronitrobenzene, as Brassicol at a dose comparable to 400 g/m<sup>3</sup>, mixed thoroughly with the soil of each pot;

f. C = control, therefore untreated soil.

The soils were mixed, freed from debris by sieving, and before treatment filled into 2½ liter plastic pots with a bottom hole for water drainage. There were 6 replicate pots for each treatment/plant combination; 3 of them received fertilization with a normal nitrogen dose (–N) and the other 3 received double the amount of nitrogen (+N). The normal fertilization was a monthly 20 ml Hoagland solution per kg of soil; the Hoagland solution comprised 30 g N.P.K. 12:10:18 + 26.65 g superphosphate + 40 g ammonium nitrate + 5 g magnesium sulphate + 0.35 g zinc sulphate + 0.075 g borax + 0.35 g copper sulphate + 1.4 g manganese sulphate + 0.075 g iron chelate in 10 liters of water solution.

The fore-mentioned 4 soils, all receiving the 5 PSS treatments described, and half of the replicates provided with a double nitrogen dose, were sown or planted with 3 different test plants 3–7 weeks after treatment and aeration, when the danger of phytotoxicity was considered to have become negligible as tested with garden cress, *Lepidium sativum* L., as an indicator plant.

The test plants sown and planted are listed below:

a. Carrot, *Daucus carota* L., about 300 seeds per pot, later thinned in three steps to 15 plants per pot;

- b. Rose, *Rosa canina* L., about 50 seeds per pot, later thinned in 3 steps to 5 plants per pot;
  - c. Ryegrass, *Lolium perenne* L., about 100 seeds per pot, left without thinning.
- The results for the different test plants are recorded and discussed under 7.2, 7.3 and 7.4, with a discussion of the results for the whole chapter under 7.5.

## 7.2. EFFECT ON CARROT

The soil was examined for nematodes before planting and after treatment, and again after carrot had been grown. The results are summarized in Table 29. Carrot seeds were sown in the treated soil on 1.12.1966, 300 seeds spaced evenly over the surface of the pot containing  $2\frac{1}{2}$  liter soil. Germinating seeds were counted and removed except for 15 plants. Germination in the PD soil was poor and the carrot was resown on 13.1.1967. The plants grew well and were finally evaluated on 29.5.1967. The results of the plant evaluation are recorded in Table 29 and in Figs 20 and 21.

Table 28 shows, that H, CP, D1 and D2 have originally killed nearly all nematodes, plant parasites as well as saprozoites, and that PCNB was not different from the untreated C. After the test crop had been grown the plant parasites were still negligible for H, CP, D1, and D2 in all four soils. The saprozoic nematodes however, had established or re-established their populations in all treatments of all soils. The very poor S soil had built up a measurable population (with PCNB as the lowest density). In all the other soils the saprozoic populations were higher in the treated than the untreated soils, except for PCNB. The quick recovery and high density reached in most treated soils is remarkable.

It is clear from Table 29 that the treatment caused significant differences in plant weight, root weight, root percentage, quality of harvested carrots and, notably, percentage of germinating seeds. Most of these differences, however, are in the order of 10% or smaller, the main difference being due to the negative effect of PCNB which had obviously been toxic in the amount applied.

The differences between treatments cannot at any rate be correlated with nematode damage or other diseases (except perhaps for the PD soil). It is probable that the small differences observed between treatments are mainly related to phytotoxicity and fertility influences, as far as they are not due to experimental variability.

Fig. 20 shows, that the S was considerably less fertile than the F and M soils. PD soil is not directly comparable to them because here the carrots were resown. Scrutiny of the results per soil indicates that extra nitrogen had a positive influence on carrot weight in the PD soil and, unexpectedly, a negative influence in the river sand S, whereas the other two soils were not responsive to extra nitrogen. There is further little continuity in the effect of the different treatments when different soils are compared. Generally the weight decreases in the order CP, D1, H, C, D2, PCNB. In soil S, however, H is highest followed by CP, in

TABLE 28. Nematode densities per 100 ml of soil in four soils treated in 5 different ways, and then sown with carrot on 1.12.1966.  $P_i$  = initial density after treatment and before planting,  $P_f$  = final density at the end of the experiment on 29.5.1967. The figures are numbers of stylet-bearing nematodes, with numbers of saprozoic nematodes between brackets; all figures are averages of 4 replicates for  $P_i$  and of 6 replicates for  $P_f$ .

Treatments	Soils		S		F		M		PD	
	$P_i$	$P_f$	$P_i$	$P_f$	$P_i$	$P_f$	$P_i$	$P_f$	$P_i$	$P_f$
H	0 (1)	-	0 (635)	-	0 (15)	-	8 (3124)	-	0 (279)	-
CP	0 (4)	-	0 (386)	-	0 (2)	-	3 (3768)	-	0 (2)	-
D1	0 (2)	-	0 (511)	-	0 (31)	-	8 (3484)	-	2 (11)	-
D2	0 (3)	-	0 (214)	-	0 (21)	-	9 (1472)	-	1 (5)	-
PCNB	0 (68)	-	0 (47)	-	5 (1250)	-	12 (1467)	-	100 (1650)	-
C	0 (15)	-	0 (388)	-	15 (970)	-	3 (1091)	-	211 (1295)	-
									3348 (1144)	-
										503 (1708)
										1812 (1724)

soil F it is D1, or even C, followed by CP, in soil M it is CP, followed by D2, and in soil PD it is CP, followed by H and D1.

Two growth criteria, however, deserve further attention, because they indicate special carrot problems for some if the soils and a PSS effect on it.

Firstly the weight percentage of second quality carrot is significantly higher for C, PCNB and D2. This means in fact that H, CP and D1, had improved the quality of the carrots considerably, although the total weight had improved only slightly. Close examination of Table 29 shows that the effect is mainly due to PD soil, where 33% of the carrots were of second quality, as against 3–5% for the other soils. It is probable that the poor quality was due to a disease.

TABLE 29. Evaluation of carrot, grown in four soils treated in 5 different ways and furnished with two fertilizer levels (cf. text for explanation of corresponding indication symbols). All figures are averages of 6 replicate pots. L.S.D. = least significant differences at 5% level, N.S. = not significant, \* = significant at 5% level, \*\* = significant at 1% level. The soils were treated on 10.10.1966, carrot seeds were sown on 1.12.1966, approximately 300 seeds per pot with 2½ liter soil; germinating seeds were counted and removed except for 15 final plants. Final evaluation took place on 29.5.1967.

Plant growth criteria	Plant weight in g.	Root weight in g.	Root as % of plant weight	Weight % second quality carrots	Excess number of germinated seeds, which were removed
<i>A. PSS treatment averages</i>					
H	155	91	58	8	178
CP	166	100	60	4	224
D1	160	102	63	7	157
D2	154	96	61	15	63
PCNB	137	81	58	20	69
C	155	97	62	15	87
L.S.D.	10.7	5.6	1.9	10.3	9.6
<i>B. Nitrogen level averages</i>					
+ N	156	94	59	12	129
– N	153	95	62	11	131
L.S.D.	N.S.	N.S.	1.1	N.S.	N.S.
<i>C. Soil type averages</i>					
S	125	66	51	3	162
F	176	106	60	5	124
M	185	122	66	5	81
PD	132	84	64	33	153
L.S.D.	8.7	4.6	1.6	8.4	7.9
<i>D. Significance of interactions</i>					
A × B	N.S.	N.S.	*	N.S.	N.S.
A × C	N.S.	*	**	N.S.	**
B × C	**	**	**	N.S.	N.S.

According to Table 28 this cannot have been nematode damage. It is possibly connected with the second problem.

The second problem concerns germination and emergence of the seed. 300 seeds were sown evenly spaced per pot, and finally 15 plants were kept whereas the others were removed as excess. The excess figures in Table 29 show, that the number of seedlings present was much higher for CP, H and D1, than for

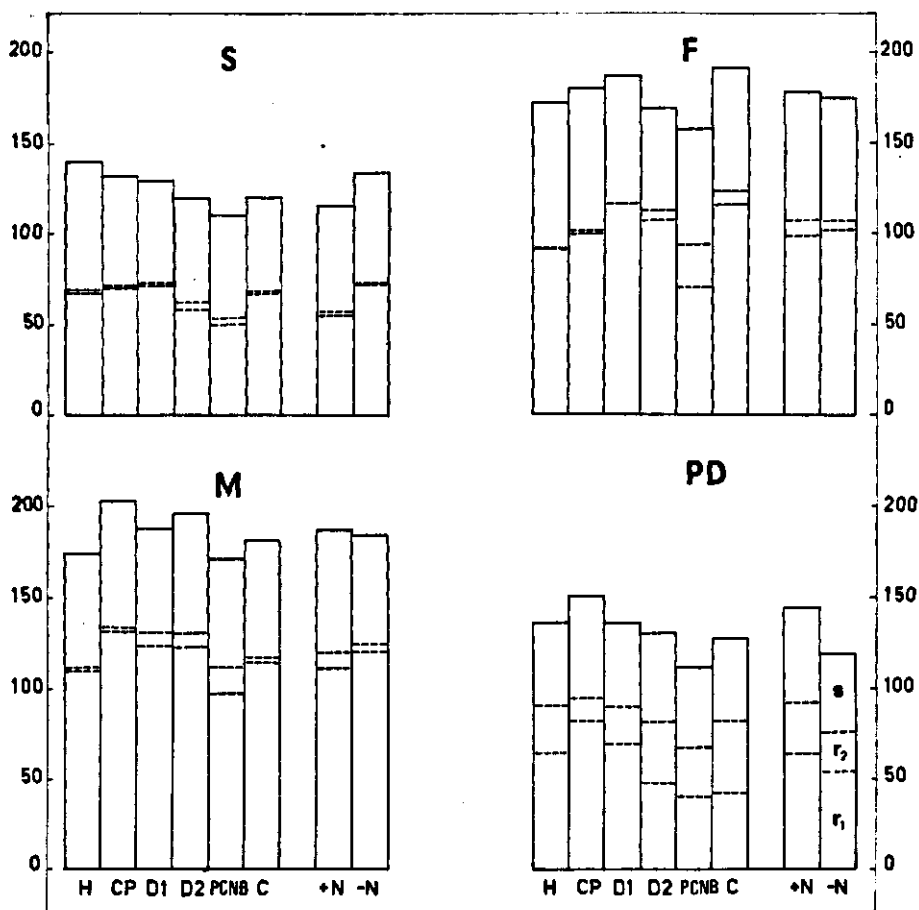


Fig. 20. Weight of carrot grown in 2.5 liter pots with four different soils (S, F, M and PD) following different PSS treatments (H, CP, D1, D2, PCNB and C), each at two different nitrogen levels (+ N and - N); cf. text for explanation of the letters. H, CP, D1, D2, PCNB and C are the average results of 6 replicate pots irrespective of nitrogen level; each column + N and - N is the average result of 18 replicate pots irrespective of soil disinfection treatments. Abscissa for each soil diagram: indication of soil disinfection treatment (at left) and of nitrogen level (at right). Ordinate for each soil diagram: weight of carrots per pot in g, subdivided in first class roots (r1), second class roots (r2) and shoots (s).

the untreated C, whereas the numbers for D2 and PCNB were even somewhat lower. This, however, varied strongly with soil type, and the interactions between treatment and soil type are also highly significant.

Taking into account some other data as well, the following conclusions can

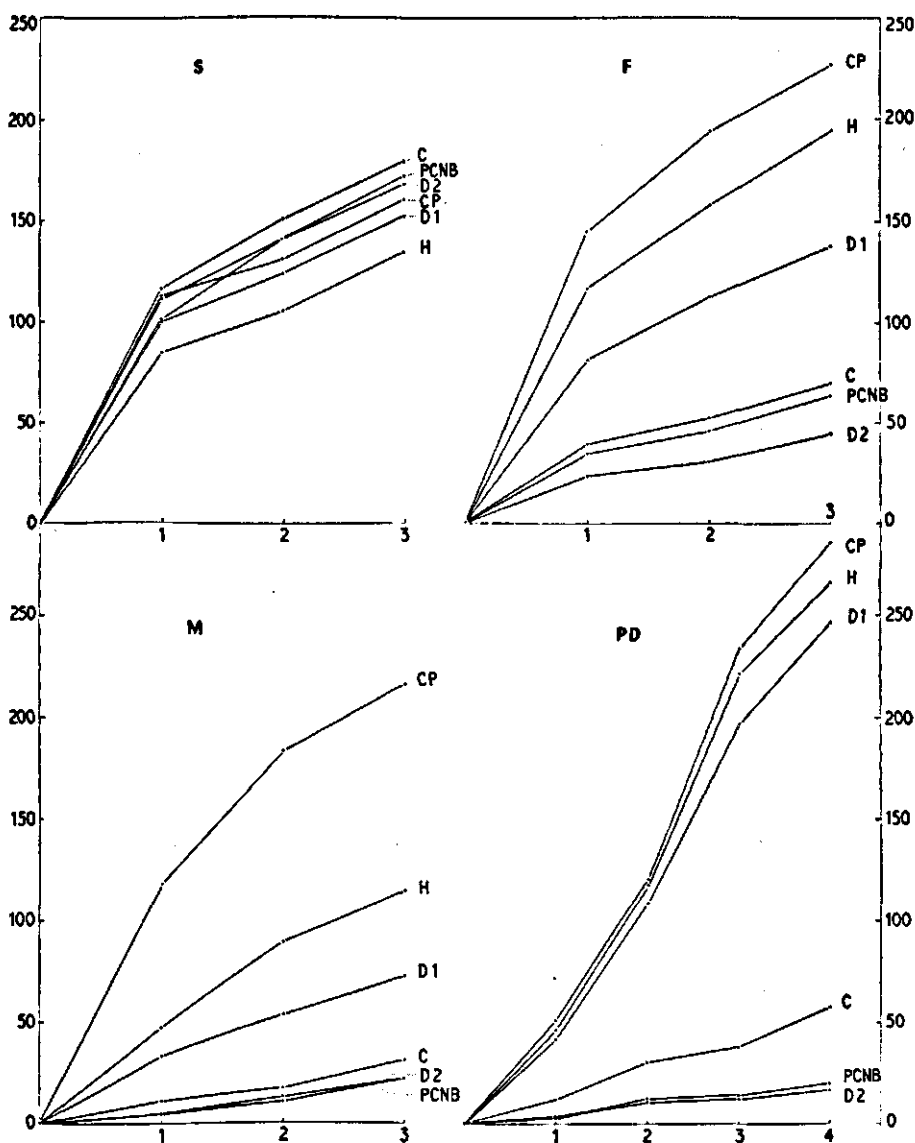


FIG. 21. Clipped seedlings of carrot grown in 2.5 liter pots with four different soils (S, F, M and PD) following different treatments (H, CP, D1, D2, PCNB and C), cf. text for explanation of the letters. Results are averages of 6 replicates. Abscissa for each soil: indication for carrot thinning times. Ordinate for each soil: cumulative number of clipped seedlings per pot.

be made: S, the pure river sand, does not have the problems the seeds germinated and emerged well and the treatments had no effect.

For F, the sandy polder soil, CP, H and D1 strongly increased the number of seedlings, although decreasing in this order. PCNB and D2 decreased the number of seedlings compared to control. The same was true for M, the old meadow soil. The problem was most marked for PD soil. Here the numbers of emerged seedlings for D2 and PCNB, and also C, were so low that the whole series was re-sown on 13.1.1967. However, it did not change the situation: there was excellent germination and emergence in the CP, H and D1 soils whereas they were poor in the untreated C and even poorer in the PCNB and D2 soils. Early counts, 12 days after sowing, already showed the numerical differences and proved that it was not in the first place due to disappearance of seedlings after emergence.

The difference could be due to differences in germination, or to differences in survival and emergence of the germinated seeds.

Observation showed that many seeds germinated and died before emergence. Mycological observation showed that seedlings in the soils with poor emergence mostly carried *Pythium* spp., which were not present in the soils with good emergence (although *Mucor* and other species occurred there). The differences in emergence are therefore probably due to fungus, perhaps *Pythium* infestations. It remains to be solved whether they were present on the seed and were suppressed in their activity in the CP, H and D1 soils, or whether they were present in the soils and were eradicated there by the CP, H and D1 treatment (D1 was apparently fungicidal whereas the lower D2 dose was not). These two possibilities are equal on the basis of our data. Carrot had been grown earlier on the PD soil. The fact that the polder soil F and the meadow soil M contained the problem whereas they had never been grown with carrot before, suggests seed-borne disease. The fact however that it did not show up in the pure river sand suggests a soil-borne problem. It is probable that the poor emergence of the seedlings and the high percentage second class carrots in the C, PCNB and D2 soils were caused by the same, or at any rate the same type of organisms, presumably fungi, because they were closely correlated with respect to occurrence and influence of treatments.

The influence of PSS on quality of test plants and on seed germination and seedling emergence are important problems which deserve more attention in general.

### 7.3. EFFECT ON ROSE

The results for rose are summarized in Tables 30 and 31 and in Figs. 22 and 23.

Table 30 shows, that the effects of H, CP, D1 and D2 on plant nematodes had again been excellent (D2 somewhat less than D1), whereas PCNB had not or hardly been effective according to  $P_t$  (just after treatment) but had nevertheless



TABLE 30. Nematode densities per 100 ml of soil in four soils treated in 5 different ways, and then grown with rose.  $P_i$  = initial density after treatment and before planting,  $P_f$  = final density at the end of the experiment 137 days after sowing, i.e. on 22.6.1967. The figures are numbers of stylet-bearing nematodes, with numbers of saprozoic nematodes between brackets; all figures are averages of 4 replicates for  $P_i$  and of 6 replicates for  $P_f$ .

Treatments	Soils		F		M		PD	
	$P_i$	$P_f$	$P_i$	$P_f$	$P_i$	$P_f$	$P_i$	$P_f$
H	0 (1)	0 (725)	0 (15)	77 (2653)	0 (279)	15 (4245)	0 (3)	121 (1580)
CP	0 (4)	0 (530)	0 (2)	64 (3198)	0 (2)	40 (3465)	4 (4)	83 (1924)
D1	0 (2)	0 (1183)	0 (31)	143 (2001)	2 (11)	50 (2312)	33 (7)	69 (2398)
D2	0 (3)	0 (603)	0 (21)	164 (793)	1 (5)	88 (1124)	19 (6)	778 (877)
PCNB	0 (68)	0 (579)	5 (1250)	87 (1132)	100 (1650)	149 (1668)	3260 (1190)	298 (1371)
C	0 (15)	0 (479)	15 (970)	29 (731)	211 (1295)	581 (1823)	3348 (1144)	1687 (1011)

TABLE 31. Evaluation of rose, grown in four soils treated in 5 different ways and furnished with two fertilizer levels (cf. text for explanation of corresponding indication symbols). All figures are averages for 6 replicate pots. L.S.D. = least significant differences at 5% level. N.S. = not significant, \* = significant at 5% level, \*\* = significant at 1% level. The soils were treated on 10.10.1966. Rose seeds were sown on 5.2.1967,  $\pm$  50 seeds per pot of 2½ liter of soil; germinating seeds were counted and removed except 5 final plants. Final evaluation took place 137 days after sowing.

Plant growth criteria	Plant length in cm.	Plant weight in g.	Root weight in g.	Root weight as % of plant weight in g.	Plant number	Excess seedlings removed
<i>A. PSS treatment averages</i>						
H	29.9	37.9	23.8	63	9	4
CP	30.6	40.5	24.6	63	8	2
D1	28.1	40.4	26.3	66	8	3
D2	27.8	41.6	26.3	66	8	3
PCNB	21.1	22.9	12.6	55	6	1
C	28.3	37.6	23.3	63	8	3
L.S.D.	4.29	4.81	2.96	2.8	1.7	N.S.
<i>B. Nitrogen level averages</i>						
+ N	29.4	37.8	22.1	59	8	3
- N	25.6	35.9	23.5	66	8	3
L.S.D.	2.48	N.S.	N.S.	1.6	N.S.	N.S.
<i>C. Soil type averages</i>						
S	9.8	13.3	9.5	69	8	3
F	30.4	40.7	23.2	56	7	2
M	39.0	50.8	28.1	54	8	3
PD	30.7	42.8	30.4	71	8	3
L.S.D.	3.51	3.93	2.42	2.3	N.S.	N.S.
<i>D. Significance of interactions</i>						
A $\times$ B	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
A $\times$ C	N.S.	**	**	**	N.S.	N.S.
B $\times$ C	*	*	**	N.S.	N.S.	N.S.

suppressed the plant nematodes to a low level after the test plant had been grown. PCNB, therefore, has also exerted a slight, but measurable nematicidal effect.

Table 31 and Figs 22 and 23 substantiate that the influence of PSS on rose in these soils had been small apart for the phytotoxic effect of PCNB, much the same as was found for carrot.

According to Fig. 22 S was again considerably less fertile than the other soils. Only for the M and PD soil the PSS treatments (except PCNB) as well as the extra nitrogen doses were somewhat favourable for plant growth when length of the plants was taken as a criterion. It is therefore probable that this slight effect was caused by nitrogen.

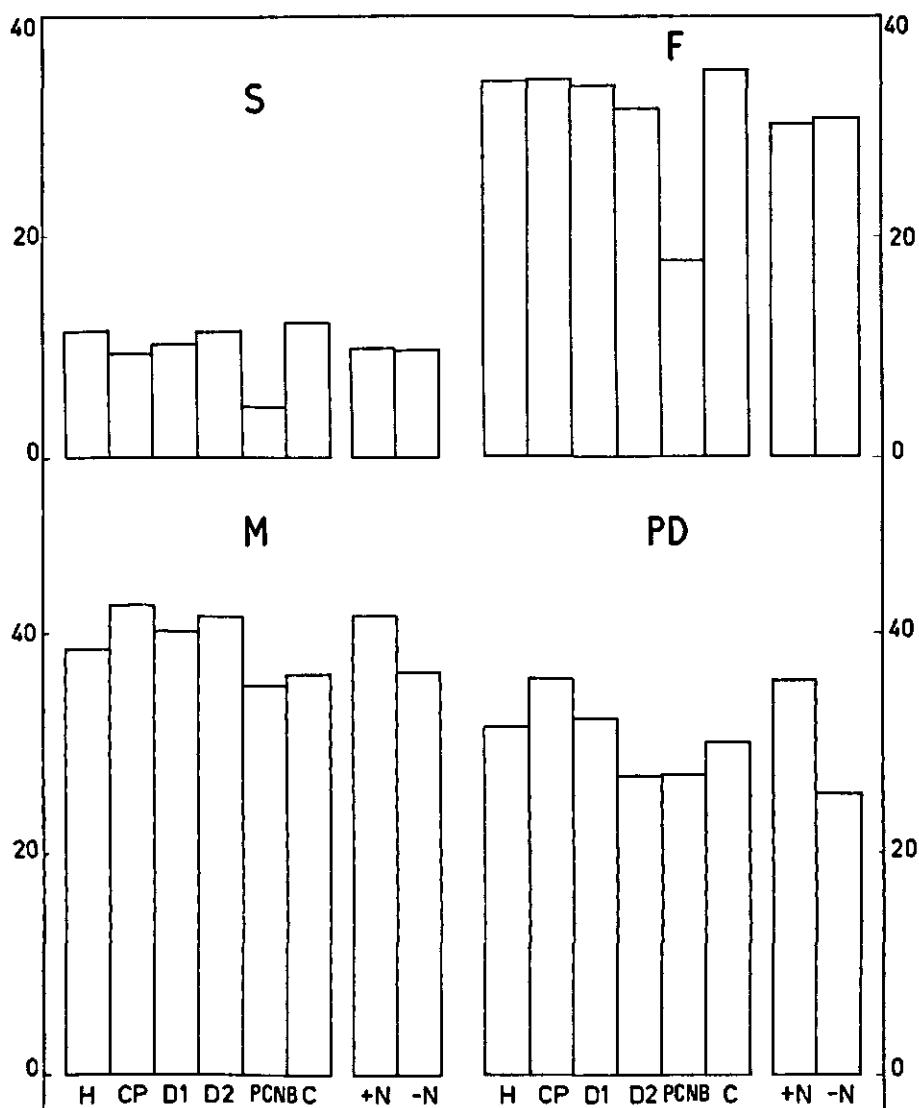


FIG. 22. Length of rose when grown in 2.5 l and pots with four different soils (S, F, M and PD), following different soil treatments (H, CP, D1, D2, PCNB and C), each at two nitrogen levels (+ N and - N); cf. text for explanation of the letters. H, CP, D1, D2, PCNB and C are the average results of 6 replicates independent of nitrogen levels; each column + N and - N is the average result of 18 replicate pots independent of soil disinfection treatment. Abscissa for each soil diagram: indication of soil disinfection treatment (at left) and nitrogen level (at right). Ordinate for each soil diagram: shoot length of rose plants in cm.

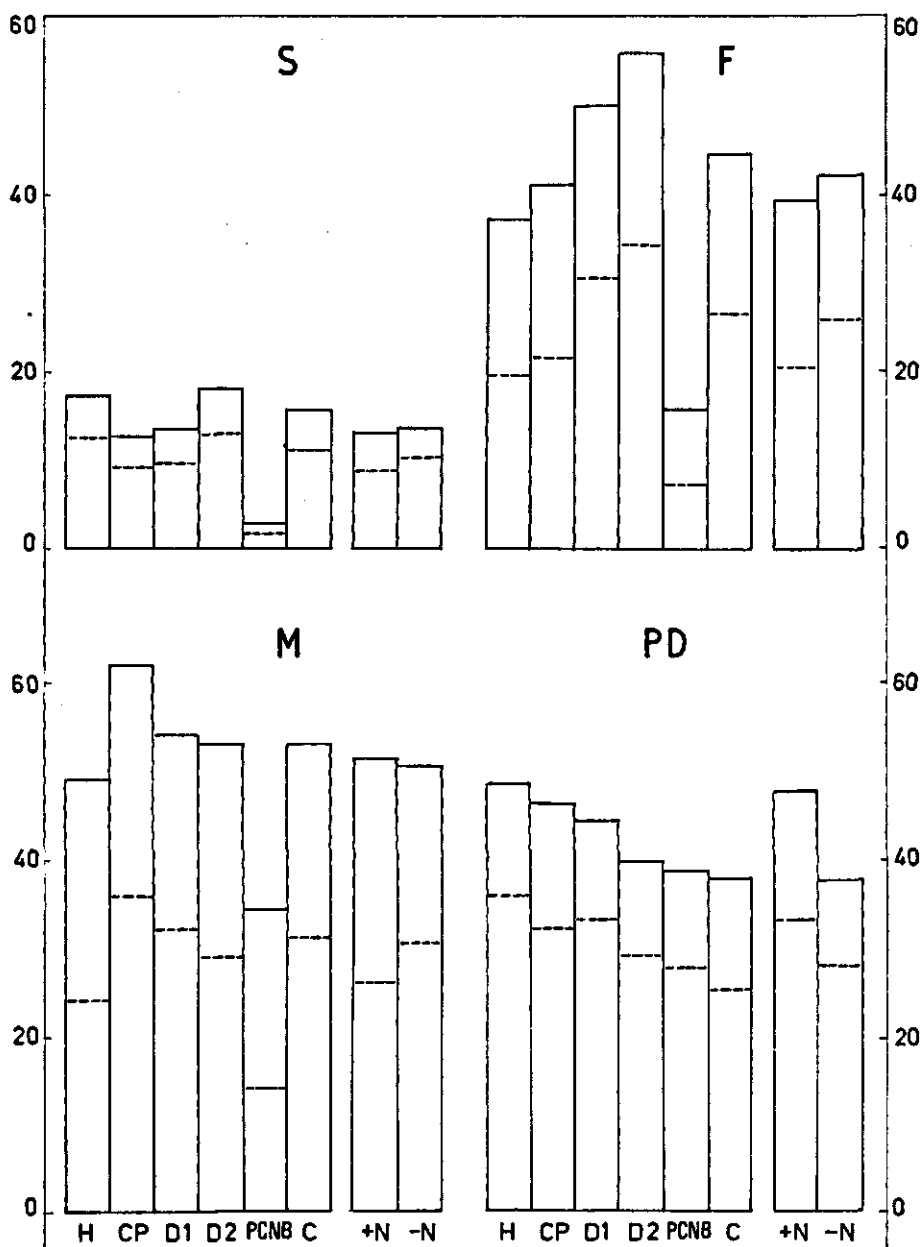


FIG. 23. Weight of rose when grown in 2.5 l pots with four different soils (S, F, M and PD), following different soil treatments (H, CP, D1, D2, PCNB and C), each at two nitrogen levels (+ N and - N); cf. text for explanation of the letters. H, CP, D1, D2, PCNB and C are the average result of 6 replicate pots independent of nitrogen level; each column + N and - N is the average result of 18 replicate pots independent of soil disinfection treatment. Abscissa for each soil diagram: indication of soil disinfection treatment (at left) and nitrogen level (at right). Ordinate for each soil diagram: weight of rose plants in g per pot, subdivided in roots (at bottom) and shoots (on top) respectively.

Fig. 23 indicates that plant weight showed some abnormal fluctuations, which differed with soil type and which may be related to manuring and toxic effects, as indicated by treatment-soil type and nitrogen level-soil type interactions (cf. Tables 29 and 31), but cannot be fully explained. However, for rose, they do not indicate the presence of a marked yield deficiency due to disease in any of these soils. It is noteworthy that the number of excess seedlings and therefore emergence of the seedlings, was not influenced by the treatments, as for carrot which seems to be seriously affected in its early stage by a mycological factor.

#### 7.4. EFFECT ON RYEGRASS

The treatment effects on nematodes as well as on ryegrass weights (separately for 5 successive cuts and for the total) are summarized in Table 32 and Fig. 24, specifically for each soil.

Table 32 indicates, like for the experiments with carrot and rose, that all treatments except PCNB have eliminated the plant nematodes effectively. D2 has been somewhat less effective than D1 in PD soil, or has at least allowed *Paratylenchus* to build up a fairly great density when considering  $P_f$ . PCNB was not effective immediately after the treatment when considering  $P_i$ , but the final density  $P_f$  was low compared to control and it must have exerted a slow nematocidal effect.

Fig. 24 shows, that the effects of the treatments were significant, but small for all soils. On the other hand, the effect of extra nitrogen was highly and significantly positive for all soils (although interactions between soil type and nitrogen were present), and most of the differences observed in this experiment may be nitrogen effects. PCNB was also hardly or not at all phytotoxic, like for carrot and rose. The following comments can be made for the different soils. For S, the river sand, treatment effects were negligible, whereas nitrogen effect was marked.

For F, the Flevoland soil, H and CP were somewhat better than the other treatments, which hardly differed from the untreated control. Nitrogen effect was very great, but it only became obvious after the third cut.

For M, the old meadow soil which was dried before use, the effects were similar as for F, but the nitrogen effect became obvious after the second cut. It also appeared that after the first and second cuts C and PCNB yielded considerably less than the other treatments. This difference disappeared later.

For the PD soil H, CP, D1 and originally also D2, were superior to PCNB and C. Later on D2 fell behind H, CP and D1. This may be related to the strong increase of *Paratylenchus* sp., particularly in this treatment. The total effects for this soil, however, are relatively small as for the other soils and may also be connected to the strong response to nitrogen in this soil.

The conclusion from this experiment is therefore, that PSS did not increase the yields much on any of these soils, although H and CP, and to a lesser extent D, showed some effect. PCNB was slightly phytotoxic to grass only in the first cuts. The effect of N was great on all four soils.

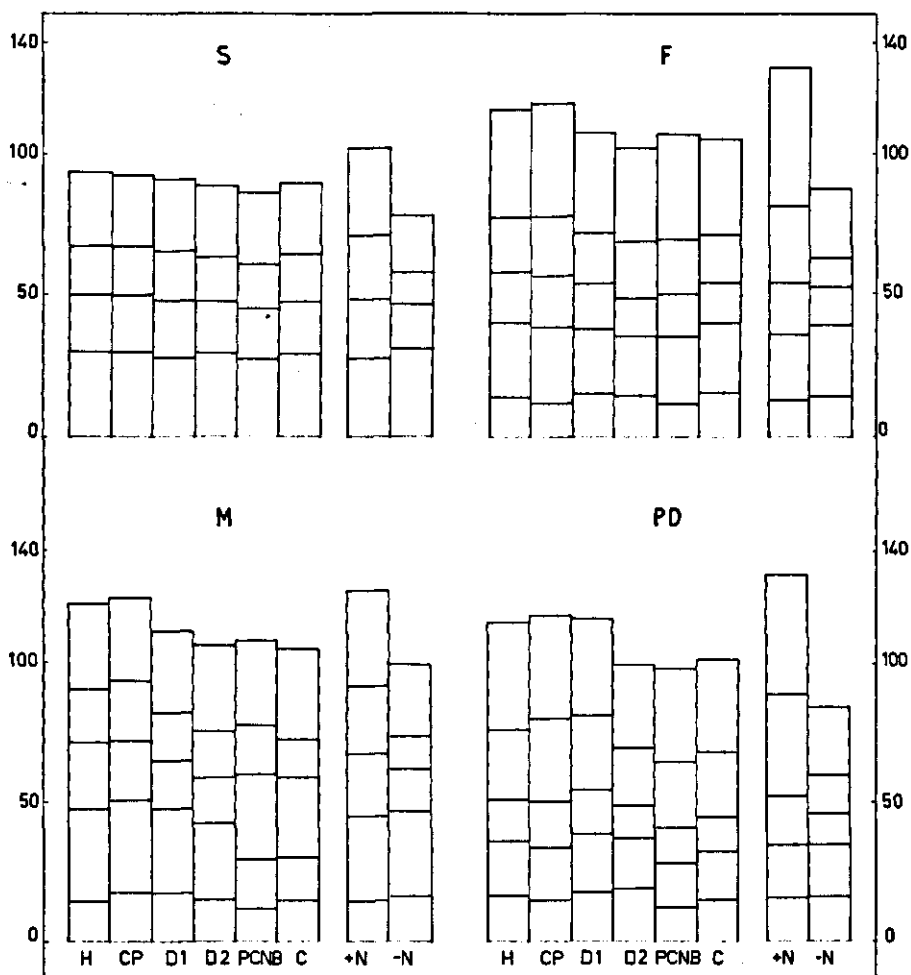


FIG. 24. The successive cuts of grass grown in 2.5 l pots with four different soils (S, F, M and PD) following different soil treatments (H, CP, D1, D2, PCNB and C) each at two nitrogen levels (+ N and - N); cf. text for explanation of the letters. H, CP, D1, D2, PCNB and C are the average results of 6 replicate pots independent of nitrogen level; each column + N and - N is the average result of 18 replicate pots independent of soil disinfection treatment. Abscissa for each soil diagram: indication of soil disinfection treatment (at left) and nitrogen level (at right). Ordinate for each soil diagram: weight of grass per pot in g, subdivided in the successive cuts respectively from bottom to top 1, 2, 3, 4 and 5 (for S: 1 + 2, 3, 4 and 5).

TABLE 32. Nematode densities per 100 ml of soil in four soils treated in 5 different ways, and grown with rye grass.  $P_i$  = initial density after treatment and before planting,  $P_f$  = final density at the end of the experiment 365 days after sowing, i.e. on 22.11.1967. The figures are numbers of stylet-bearing nematodes, with numbers of saprozoic nematodes between brackets; all figures are averages of 4 replicates for  $P_i$  and of 6 replicates for  $P_f$ .

Treatments	Soils		S		F		M		PD	
	$P_i$	$P_f$	$P_i$	$P_f$	$P_i$	$P_f$	$P_i$	$P_f$	$P_i$	$P_f$
H	0 (1)	-	0 (1218)	-	0 (15)	13 (1268)	0 (279)	-	0 (3)	36 (1791)
CP	0 (4)	-	0 (404)	-	0 (2)	8 (1948)	0 (2)	-	4 (4)	28 (1495)
D1	0 (2)	-	0 (94)	-	0 (31)	18 (1844)	2 (11)	-	33 (7)	11 (2353)
D2	0 (3)	-	0 (175)	-	0 (21)	11 (906)	1 (5)	-	19 (6)	318 (1815)
PCNB	0 (68)	-	0 (176)	-	5 (1250)	37 (724)	100 (1650)	-	3260 (1190)	150 (573)
C	0 (15)	-	0 (207)	-	15 (970)	14 (490)	211 (1295)	-	3348 (1144)	1364 (2696)

## 7.5. DISCUSSION

The results in Tables 27-32 and Figs 20-24 show that PSS had very little influence on plant growth in river sand, pathogen-free Flevoland soil, and meadow soil which had previously been exposed to drought. The effects in PD soil were also small for carrot, rose and grass (which had never been grown in it before), but were greater than for the other soils. The effects which are usually obtained in old cultivated soils (cf. Chapters 4-6) are much greater and therefore cannot be due to direct influence of the chemicals on plant growth, nor also to the nitrogen effects, because an excess of nitrogen was usually applied in these experiments.

The nitrogen factor nevertheless requires further discussion. Addition of a double dose of ammonium nitrate increased the growth of grass in treated soils of all types, but the responses of rose and carrot were more complicated. Growth of rose was promoted by extra nitrogen in PD soil and perhaps in M soil, but it was decreased in F soil and not influenced at all in S soil. Growth of carrot was promoted in PD soil, decreased in S soil and hardly influenced in F and M soils. This reflects the knowledge already cited in the literature review, that the nitrogen effects of PSS may as often be favourable as unfavourable to plant growth, depending on the soil and the test plant used. PSS effects recorded in Chapters 4-6 must have been influenced by this phenomenon, but they are strong and consistent and most of these effects must be due to other causes than nitrogen effects according to the figures in this Chapter.



## 8. DIAGNOSTIC STUDIES ON AN UNIDENTIFIED EFFECT IN OAT

### 8.1. INTRODUCTION

PSS has demonstrated several unidentified problems of poor growth in the foregoing inventory of experimental results. Severe yield deficiencies may be expected to show up if soils infested with a noxious pest or disease are treated (cf. Chapter 6). A number of marked problems, however, cannot be explained on this basis and require further diagnostic study. Poor growth of oat is one of them and was selected for further study.

Cereals are grown on more than half of the cultivated land in The Netherlands (FEEKES 1967). They show marked yield deficiencies on sandy and peaty soils. This is obvious from Tables 2 and 3 for oat and barley, which reached an average 134%/145% and 124%/147% on disinfested soil as compared to untreated. Such a yield loss occurred on soils where major nematode pests, such as *Heterodera avenae*, or other noxious parasites could not be located. Table 6 shows, that PSS caused a yield increase for oat up to 237% in soils where the nematode complex *Pratylenchus crenatus* + *Tylenchorhynchus dubius* + *Rotylenchus robustus* (nr. 5 in Table 5) was present. Poor growth of oat was also shown to be negatively correlated with the preplant density of this complex in a field trial (OOSTENBRINK 1966). Negative correlations between the growth of cereals and the densities of each of these species are also recorded (OOSTENBRINK 1959; DECKER 1963 & SHARMA 1971).

It is, however, obvious from Tables 2 and 3 and from the pot experiments recorded under 6.9, that broad-spectrum soil disinfectants were often more effective than nematicides, whereas Captan was also effective in some soils. Furthermore the problem has not been fully reproduced by inoculation with nematodes or other organisms. For these reasons further experiments were made with PD-garden soil and with oat as a test plant.

### 8.2. EXPERIMENTS

#### 8.2.1. Introductory experiment 1968

Some selected PSS treatments were applied to PD-garden soil, whereas simultaneously another amount of the same soil was heated and then inoculated either with undifferentiated soil extracts or with nematodes. PD-garden soil is a sandy loam, the granular composition of which has already been recorded in Table 27. After passage through a coarse sieve and thorough mixing the soil was placed in square, plastic containers of  $4 \times 4 \times 20$  cm, containing about 300 ml = 350g of soil each; the bottom had openings for drainage and two plastic straws were inserted for good aeration. After mixing, but before treatment, the nematode

population per tube comprised notable densities of *P. crenatus*, *T. dubius*, *R. robustus*, and also *Trichodorus pachydermus*, other stylet-bearing nematodes and saprozoic nematodes; cf. Table 33.

Nematodes and other inoculation materials were washed from portions of this soil by elutriation and caught on a stack of four sieves with a  $44\ \mu$  mesh. Part of the catch was placed on cottonwool filters, which allowed the nematodes to pass into clean water. This furnished soil-free nematode suspensions which were washed in tap water. Another part of the catch on the sieve including small soil particles, organic debris and fungus hyphae and spores (ANONYMOUS 1968, GARRETT 1964, PARKINSON & WILLIAMS 1961 and MCCAIN et al. 1967) was also used as inoculum.

The following treatments were applied to the naturally infested soil:

1. C = untreated control soil comprising the normal, natural density (N) of soil organisms;
2. H = soil heated at  $60^{\circ}\text{C}$  for 2 hours;
3. CP = chloropicrin 0.25 ml/l of soil, comparable to  $50\ \text{ml/m}^2$ . The soil was placed in sealed plastic bags for 7 days, after which aeration for 10 days was allowed before sowing;
4. D = dichloropropene in the formulation of DD, applied at the rate of 0.15 ml/l of soil, comparable to  $30\ \text{ml/m}^2$ ; the soil was treated as for CP;
5. T = Temik, with 10% methyl-(methylthio) — propionaldehyde — (methyl carbamoyl)-oxime as active material, 0.025 g/l (comparable to  $5\ \text{g/m}^2$ ), placed directly under the seed;
6.  $\frac{1}{4}\text{C} + \frac{3}{4}\text{H} = \frac{1}{4}$  untreated soil +  $\frac{3}{4}$  heated soil;
7. C + 3S = untreated control soil + the uncleaned soil extract (S) of 3 times the same amount of soil (cf. Fig. 25);
8.  $\frac{1}{4}\text{N} =$  heated soil +  $\frac{1}{4}$  of the normal nematode density of the untreated control soil;
9. N = heated soil + normal nematode density of the untreated control soil.
10. 4N = heated soil + 4 times the normal nematode density of the untreated control soil.

Ten replicate containers for each treatment were grown with oat, *Avena sativa* 'Marne'. One germinated seed was planted per container on 22.7.1968. Some seeds in the C and C + 3S containers, which generally showed poor growth throughout the experiment, died early and were replanted. All tubes were fertilized after 10 days with Hoagland solution, 6 ml per tube. Water was applied at regular intervals. Plant growth showed great differences between the differently treated objects, and growth was evaluated. After 94 days the plants were harvested, counted, measured and weighed. The soil samples and root samples were extracted for nematode counts. The results are summarized in Table 33.

It appears from the table that the plants in the control soil (1) had developed much poorer than in the soils treated with heat, chloropicrin or DD (2, 3 and 4). This was visible in all growth statistics, which were apparently well correlated. The differences between the successful treatments 2, 3 and 4 were not significant

TABLE 33. Development of oat and final densities of nematodes when PD-garden soil was placed in tubes of 300 ml and treated in ten different ways, as indicated. Sowing date 22.7.1968, evaluation date 24.10.1968. Figures are absolute figures per tube and are averages of ten replicates, unless indicated otherwise. S = the undecanted extract from 300 ml control soil, N is the catch of active nematodes from the same amount of soil. The normal, initial number of nematodes per 300 ml of soil before treatment was: 1125 P. (= *Pratylenchus crenatus*) + 900 T. (= *Tylenchorhynchus dubius*) + 660 R. (= *Rotylenchus robustus*) + 135 Tr. (= *Trichodorus pachydermus*) + 615 O. (= other stylet-bearing nematodes) + 4230 S. (= saprozoic nematodes). L.S.D. = least significant differences at 5% level.

Plant and nematodes Treatments, cf. text	Root weight in g	Shoot weight in g	Shoot length in cm	Num- ber of tillers	% tubes with flowering plants	Final numbers of nematodes, logarithmically transformed					
						in roots: P. per 10 g roots		in soil:			
						P.	T.	R.	Tr.	O.	S.
1 = C (control)	0.33	1.55	36	2.9	20	3.77	2.99	2.44	0.31	2.17	3.66
2 = H (heated)	1.81	7.46	90	3.6	90	0.00	0.00	0.00	0.00	0.00	3.63
3 = CP (chloropicrin)	2.18	9.77	89	3.2	100	0.00	0.00	0.00	0.00	0.00	3.53
4 = D (DD)	2.09	7.72	80	3.3	100	0.00	0.00	0.00	0.00	0.00	3.75
5 = T (Temik)	0.76	3.55	56	1.7	50	0.66	1.08	0.86	0.00	1.08	2.63
6 = 1/4 C + 3/4 H	1.67	11.84	96	3.1	90	2.91	3.13	1.95	0.00	1.96	3.92
7 = C + 3 S	0.16	0.83	36	2.4	20	3.27	2.37	0.86	0.08	1.80	3.72
8 = 1/4 N	1.45	10.04	87	3.7	60	1.78	3.11	1.48	0.00	1.30	3.84
9 = N	1.42	8.03	87	2.6	100	2.52	3.54	2.32	0.10	1.63	3.75
10 = 4N	1.36	6.83	93	2.1	100	3.17	3.86	3.20	0.34	1.31	3.86
L.S.D.	0.48	3.39	16	1.2	29	0.326	0.277	0.360	N.S.	0.377	0.234

in any yield statistic. There was a favourable effect of Temik as well, but on growth the difference with the control was smaller and significant only in shoot length. This may be connected with a phytotoxic effect, due to the fact that Temik was placed directly under the seed and not applied in advance as with the other treatments. It appears further that inoculating the purified nematodes into heated soil (8, 9 and 10) had little effect on growth of the test plant as compared to the heated soil (2), except for root weight which was suppressed. Plant growth was also not noticeably reduced when  $\frac{1}{4}$  control soil was added to  $\frac{3}{4}$  heated soil (6). This was quite different for object 7, in which control soil was inoculated with soil extract S from 3 times the same amount of control soil. In this object plant growth was as poor as in the control soil (1); root and shoot weights were even lower, but these differences were not significant.

A mixed nematode population, with *P. crenatus*, *T. dubius* and *R. robustus* most numerous, was present before treatment. All three species were still numerous in the control soil after growth of the test plant oat, although *Pratylenchus* and *Rotylenchus* had somewhat decreased in numbers. In the soils treated with heat, chloropicrin and DD the plant parasitic nematodes were evidently eradicated, for none were found even after the growth of the test crop, whereas the saprozoic nematodes were again about as numerous as in the control soil. Temik had also reduced the population markedly, but some plant parasitic nematodes were left. The number of saprozoic nematodes after the test crop was much lower than for any other object indicating a long-lasting nematicidal effect of Temik. Object 6 ( $\frac{1}{4}$  control soil +  $\frac{3}{4}$  heated soil) shows that inoculation of 1 part infested soil with 3 parts heated soil is not sufficient to cause disease symptoms, and therefore that the degree of damage depends on the preplant density of the noxious agent. It also shows that *T. dubius* reproduced much faster than the other prevailing species *R. robustus* and *P. crenatus*. Object 7 is the most interesting object. It comprised fewer plant parasitic nematodes than object 1, with which it was comparable apart from the fact that soil extract, 3S, was added to object 7. The addition of the soil extract has obviously led to suppression of the original nematode population and not to an increase. The nematode density was generally even lower than for 6. This is probably due to the poor growth of the test plant which itself must have been induced by the soil extract. The pots inoculated with increasing densities of cleaned nematodes showed a higher final density of *T. dubius*, and at the highest inoculum also for *R. robustus*, but not for *P. crenatus*. *Pratylenchus* is again much lower than for the control soil; this holds for the soil as well as for the roots, even in the 4N objects. The inoculation of cleaned *Pratylenchus* has apparently led to a defective infection, although the test plant developed properly in these objects.

It is obvious, therefore, that the soil harboured a serious disease for cereals, which was cured by PSS. It was fully reproduced by inoculation of uncleaned soil extract and only partially by inoculation of nematodes. This result would suggest that an agent other than the nematodes caused most of the damage. On the other hand: nematode infection has, at any rate for *P. crenatus*, been defective; *P. crenatus*, *T. dubius* and *R. robustus* were the only known pathogens

present in high densities and reproducing on oat; and the fact that a low dose of DD was as effective as heat or chloropicrin also indicated that nematodes were probably involved. The experiment, therefore, was inconclusive. Final nematode densities were assessed exactly, but these reflect not only the original rate of infection which determines the damage, but also the development of the test plant which determines the rate of nematode reproduction to a larger extent. The problem is studied further in the following experiments.

#### 8.2.2. Experiments in 1969

Four inoculation experiments were made in 1969 to analyse the disease complex. The same materials were used as in 1968, namely the oat variety 'Marne', PD-garden soil, and plastic containers of  $4 \times 4 \times 20$  cm. The tubes were, however, now filled with 50 ml of river sand at the bottom, 300 ml of the test soil on top of it, and finally a thin layer of gravel to protect the soil structure. Water was provided regularly, the Hoagland solution (6 ml per tube) was added once a month, whereas 0.5 g Dolocal was added during the growing season.

The inocula used in the experiments were collected from the naturally infested soil according to the scheme of Fig. 25. They are: the whole population of active nematodes extracted from the soil into clean water (N), the uncleaned soil extract from the collector sieves without further treatment (S) and after strong blending and drying (F), the loose root pieces in various stages of deterioration retained from the soil on the top sieve of the elutriator without further treatment (R) and after heating (HR), and also active *Pratylenchus* specimens extracted from the loose root pieces (P).

The inocula in the various experiments were applied to heated soil (H). Four inoculum levels were always used, namely  $\frac{1}{4}$ , 1, 4 and 16 times the normal concentration in the naturally infested soil. The naturally infested soil itself was present in all experiments as a control (C). Ten replicate tubes were available for each treatment.

Planting and evaluation dates and results are recorded for the four experiments under a-d.

##### a. Inoculation of root pieces from soil (R, HR).

The root pieces (cf. Fig. 25) were inoculated into a heated portion of the naturally infested soil from which they were collected, in the amounts R/4, R, 4R and 16R. The final results are summarized in Table 34.

Heating of the soil removed the yield deficiency as in earlier experiments. It appears that inoculation into the heated soil of a large amount of roots increased growth of oat significantly, probably owing to manurial effects. Untreated roots had generally the same effect as heated roots, although the untreated roots at the amount R/4 and 4R caused some decrease in root weight. The inoculated rootlets harboured *P. crenatus*, and the final *Pratylenchus* numbers around and in the roots of the test plant oat increased with the amounts of R, but were never high. The low *Pratylenchus* densities at the end of the

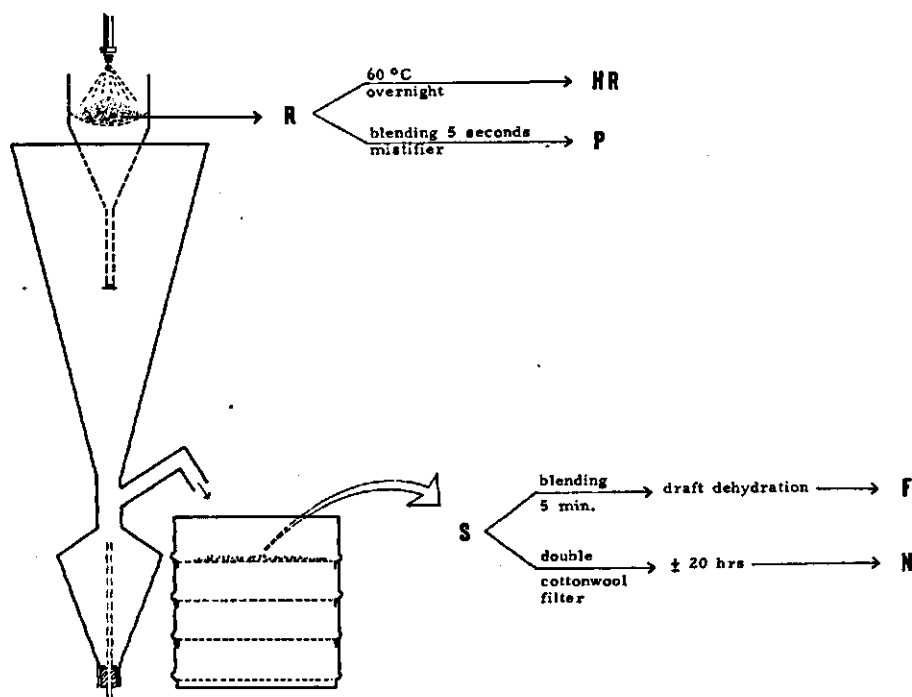


FIG. 25. Diagram showing collection procedure to obtain different materials for inoculation from infested control soil by means of the elutriator. R = loose root pieces in various stages of deterioration and caught on the 1 mm top sieve; they can be heated at 60°C overnight to form HR or they can be blended lightly for 5 seconds, and then placed in a mistifier or on a cottonwool filter to collect the *Pratylenchus* specimens, P which these root pieces still harbour. After elutriation the 44 $\mu$  collector sieves catch the uncleaned soil extract, S consisting of nematodes, other small animals and fine soil particles and organic debris, including fungi hyphae and spores. It can be blended strongly for 5 minutes and dried to kill among others, the nematodes, and then delivers F, or it can be placed on cottonwool filters in water to deliver a clean suspension of the actively moving nematodes, N.

experiment in the control C indicate, that reproduction of the nematode has stagnated. This must be due to the conditions of the soil, probably to the effect of thermal treatment, for oat is known as an efficient host.

The experiment, therefore, proves that the addition of low amounts of *Pratylenchus*-infested roots from naturally infested soil reproduced the sickness symptoms only to a very limited extent, whereas the highest amount increased plant growth, probably due to the new soil properties and decomposition of plant residues (GARRETT 1938 & 1956). The population of *P. crenatus* deteriorated during the experiment and its role cannot be judged with confidence from the results.

#### b. Inoculation of the uncleaned soil extract (S)

The uncleaned soil extract was added to a heated portion of the same soil

TABLE 34. Development of oat and final densities of nematodes when increasing amounts of root pieces were collected from nematode-infested PD-garden soil and inoculated into tubes with 300 ml of the same naturally infested soil. Amounts R/4, R, 4R and 16R. R is the normal catch from 300 ml soil, namely 3.6 g of root pieces, still comprising about 1200 *P. crenatus*. HR = heated roots. Sowing date 10.6.1969, evaluation date 26.9.1969. Figures are per tube and are averages of 10 replicates. For P., O., S. and L.S.D., cf. Table 33.

Plant and nematodes		Final numbers of nematodes, logarithmically transformed									
Treatments		Root weight in g	Shoot weight in g	Shoot length in cm	Seed weight in mg	in oat roots:			in soil:		
						P.	P.	O.	P.	O.	S.
C (control)		0.34	1.45	37	100	0.97	1.42	3.08	3.89		
H (heated & not inoculated)		0.99	1.41	38	75	0.00	0.00	0.00	3.58		
HR/4		0.66	1.12	34	99	0.00	0.00	0.00	3.57		
HR		0.96	1.56	40	121	0.00	0.00	0.00	3.74		
4HR		0.80	1.40	38	95	0.00	0.00	0.00	3.81		
16HR		1.15	1.84	44	262	0.00	0.00	0.00	3.80		
R/4		0.64	1.22	36	76	0.79	0.00	0.00	3.74		
R		0.74	1.14	37	82	1.57	0.24	0.08	3.79		
4R		0.56	1.71	41	69	1.55	0.99	0.95	3.80		
16R		2.01	3.26	49	406	2.73	1.12	1.92	3.83		
L.S.D.		0.389	0.482	7.7	147	0.396	0.368	0.320	N.S.		

TABLE 35. Development of oat and final densities of nematodes when increasing amounts of uncleanded soil extract were collected from PD-garden soil and inoculated into tubes with 300 ml of the same soil after it had been heated (S/4, S, 4S, 16 S). S is the normal extract from 300 ml soil, comprising nematodes and other fine soil particles. Sowing date 8.7.1969, evaluation date 17.9.1969. Figures are per tube and are averages of 10 replicates. For *P.*, *O.*, *S.* and L.S.D. c.f. Table 33.

Treatments	Plants and nematodes	Root weight in g	Shoot weight in g	Shoot length in cm	Final numbers of nematodes, logarithmically transformed.			
					in roots	in soil		
					<i>P.</i>	<i>P.</i>	<i>O.</i> <sup>1</sup>	<i>S.</i>
C (control)		0.99	1.40	36	2.14	1.59	2.88	3.75
H (heated, not inoculated)		1.36	1.96	40	0.00	0.00	0.00	3.77
S/4		1.73	2.56	49	1.28	0.00	2.53	3.85
S		1.09	1.96	47	1.66	0.17	3.16	3.85
4 S		0.30	1.00	31	1.70	1.60	2.39	3.65
16 S		0.20	0.43	20	0.90	2.30	2.20	3.68
L.S.D.		0.46	0.47	8.5	0.361	0.350	0.287	N.S.

<sup>1</sup> Largely *T. dubius*

from which it was collected; the amounts were S/4, S, 4S and 16S- with naturally infested soil (C) and heated soil (H) as controls. The final results are recorded in Table 35.

It is obvious that a growth deficiency problem was present, that heating the soil removed it (H), and that the inoculation of the soil extract reproduced the syndrome, as in the 1968 experiment. Heating the soil (H) caused a considerable increase in root and shoot development. The lowest inoculum amount (S/4) may have increased oat growth somewhat as compared to H, or did at any rate not decrease it. The higher inoculum amounts caused a strong growth depression. Growth figures for the amounts 4S and 16S were much lower than for H, and even much lower than for the untreated control C. These high amounts caused an extremely reduced root system with a root/shoot ratio below  $\frac{1}{2}$ , and minute but nevertheless mature and seed-producing plants, cf. Figs 26 and 27. This is apparently the ultimate state of the disease syndrome, which is more severe than occurs under natural conditions.

It appears, therefore, that growth deficiency was present in the soil and that it could be cured by heating the soil. Also that it was reproduced by adding the soil extract, that the degree of damage was related to the amount of inoculum, and that the symptoms were: serious stunting, malformation and discolouration of the roots with a consequent stunting of the aerial plant parts but not death of the plants of incomplete maturation.

The experiment, as in the similar experiment made in 1968, did not indicate that the plant parasitic nematodes were the cause of the symptoms, nor did it





FIG. 26. Roots of oat: At left normal, growing in sick soil after this soil had been heated. At right sick, growing in the same heated soil after the addition of a heavy amount of soil extract from the naturally infested soil. The sick roots are darkly coloured, short, strongly swollen, capriciously curved and densely covered with stunted root hairs.

exclude its possibility. Only final densities at the end of the experiment are available and are recorded in Table 35.

*P. crenatus*, which was present in moderate densities in and around the oat roots of the control soil, was exterminated in the heated soil and got established again proportionate to the amount of soil extract inoculated. In the object 4S, and very marked at 16S, the *Pratylenchus* specimens were apparently prevalent around and not in the oat roots. This must be due to the scarcity of roots and the unhealthy conditions of the few roots present.

The total numbers of *Pratylenchus* for 4S and 16S were about the same as for the control. They must have been depressed seriously by the very poor root development, as was the case for the other stylet-bearing nematodes. In this group, with *T. dubius* by far the most numerous species, the density for the normal amount of soil extract S was higher than for C, but increase of the inoculum to 4S and 16S did not increase the nematode density further. Instead, it was decreased to less than 20%. The low nematode figures at high inoculum densities, therefore reflect plant growth and initial infection rate at the same time, and hence do not reveal pertinent information about the initial infection alone. It is nematologically interesting to note, that the influence of heating on the density of the saprozoic nematodes was no longer visible in the final, postplant densities, but that these were positively correlated with root development.

#### *c. Inoculation of the soil extract after blending and drying (F)*

Strong blending and dehydration of the soil extract kills soil organisms which are susceptible to this rough treatment, such as nematodes and other animals, but most of the fungi spores survive. The treated soil extract was inoculated, as under b., to heated soil in amounts F/4, F, 4F and 16F, again with heated soil,

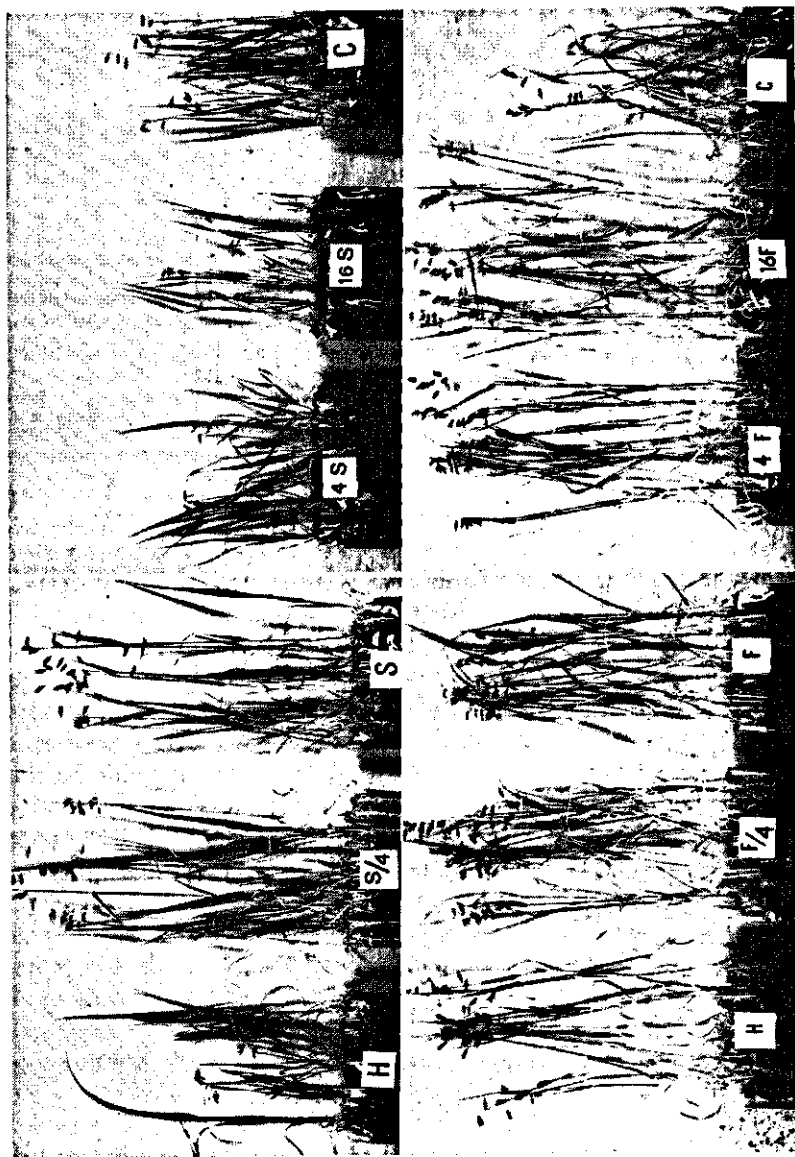


FIG. 27. Growth of the test plant oat after inoculation with increasing amounts of the untreated soil extract (S/4, S, 4S & 16S; top row of photographs) and of strongly blended and dried soil extract (F/4, F, 4F & 16F; bottom row of photographs). H is the heated soil which was inoculated; C is the untreated, naturally infested control soil, from which type of soil the soil extracts were drawn; H and C are added as controls.

TABLE 36. Development of oat and final densities of nematodes when strongly blended and dehydrated soil extract was added to heated soil (F/4, F, 4F and 16F). Sowing date 18.6.1969, evaluation date 12.8.1969; c.f. Table 35 for comparison.

Treatments	Plants and nematodes	Root weight in g	Shoot weight in g	Shoot length in cm	Final number of nematodes loga- rithmically transformed <sup>1</sup>			
					in roots	in soil		
						P.	P.	O. S.
C (control)		0.24	0.77	38	2.41	2.77	3.11	3.73
H (heated, not inoculated)		0.78	1.82	53	0.00	0.00	0.00	3.70
F/4		0.79	2.13	53	0.00	0.00	0.00	3.89
F		0.76	1.91	53	0.00	0.00	0.00	3.80
4 F		1.09	2.26	55	0.00	0.00	0.00	3.88
16 F		1.44	2.81	56	0.00	0.00	0.00	4.14
L.S.D.		0.299	0.40	4.4	0.037	0.124	0.100	N.S.

<sup>1</sup> The initial nematode densities in the control soil C before treatment were per tube P. = 2275, O. = 2730, S. = 5475, or logarithmically transformed 3.36, 3.44 and 3.74 respectively.

(H), and untreated control soil (C) as checks. The final results are recorded in Table 36.

As in the experiment under b. heating of the soil cured the problem (H versus C). The inoculation of the blended and dehydrated soil extract, caused a significant manurial effect, but did not reproduce the disease syndrome, as was the case with the untreated soil extract. The differences are clearly indicated in Fig. 27. The plant parasitic nematodes were fully controlled by the treatment of the extract. This stresses the possibility that they are involved in causing the disease. However, other organisms susceptible to the treatment of the sub-  
strat, are not excluded. The fact that the plant nematode densities in the control soil, *Pratylenchus* spp. as well as the other stylet-bearing nematodes, dropped during the experiment to about  $\frac{1}{2}$  of their original densities, indicates (as in earlier experiments) that the environmental conditions were not very favourable for them. The saprozoic nematodes increased to high numbers again in the heated soil, especially in the 16F object.

A second planting of oat, started on 13.8.1969, developed better than the first planting on the control soil, possibly due to the decline of plant nematode density during the first planting. The manurial effect of the inoculum was also gone. This explains why hardly any differences between the objects appeared. The results of this second planting, therefore, could be explained, but they did not add to further insight about the cause of the original growth deficiency problem, and the figures are not recorded here.

#### d. Inoculation of the whole natural population of actively moving nematodes (N)

The natural population of nematodes, extracted from the original, infested soil, comprises the active nematode specimens. They are elutriated, caught on

the sieves, passed onto the cottonwool filters to get rid of debris and finally of their own strength pass into water. It is the total nematofauna minus the specimens which are lost. These losses consist of immobile stages, eggs, stages in cysts (e.g. *Heterodera* spp.) or egg sacs (e.g. *Meloidogyne* spp.) or rootlets (e.g. *Pratylenchus* spp.), or specimens not passing the filters for unknown reasons (e.g. *Longidorus* and *Xiphinema* spp.). It includes an estimated 20% loss of usually active specimens due to incomplete recovery by the various manipulations in the extraction procedure. The catch, therefore, comprises some 80% of the active specimens, which may be less than half of the total nematofauna present in the soil. In our case the catch consists mainly of the *Pratylenchus* specimens – which are free in the soil – the ectoparasitic species *T. dubius*, *R. robustus* and *Tr. pachydermus*, and saprozoic nematodes. *Heterodera*, *Meloidogyne*, *Xiphinema* and *Longidorus* species were not present in this soil, but a considerable number of *Pratylenchus* specimens enclosed in the undecayed rootlets of the previous oat crop on this soil must have been lost. The same may hold for a number of eggs of all persisting species mentioned above.

The extracted mixture of active nematodes was inoculated to heated soil, as in the earlier experiments. The final results on oat growth and nematode densities, as well as the initially inoculated numbers, are recorded in Table 37.

Heating of the soil removed the growth deficiency as in the other experiments (H versus C). The nematode inoculum did suppress root and shoot weight significantly, but not shoot length. Shoot and root weights were somewhat reduced by N/4, considerably so by N, and strongly by 4N, whereas 16N was not more but even somewhat less effective than 4N. 4N nearly reached the low level of the untreated control, and considering the losses of nematodes caused by collection as well as re-inoculation, the infective strength of the inoculated amount 4N may have been similar to that of the normal, well-established population in the control soil. Inoculation of the nematodes, therefore, reproduced the damage to such an extent, that they must be considered a major cause of the growth stagnation. The nematode effect was much stronger than in the 1968 experiment (cf. Table 33). *P. crenatus* and *R. robustus* decreased again in the control soil and re-established themselves, although defectively, in the inoculated heated soils (Table 37 and footnotes). This suggests that these species are not the main parasites. It is, however, still possible that their inoculated populations caused the damage and then declined due to the general unfavourable conditions of the experiment. *T. dubius* established itself better. It was by far the most numerous species in the final population, and must have multiplied on the inoculated plants. It causes damage to cereals according to monospecific inoculations by other workers (SHARMA 1971). Therefore here it must have been noxious. Its final population densities correspond with the initial inoculum, taking into account the amount of plant roots available. It is not sure why the plants in 4N were even poorer than for 16N, but the fact that the final populations of *T. dubius*, as well as of *P. crenatus*, were correspondingly suppressed, must be due to this poor root development, as found earlier in the experiment under b. (cf. Table 35). *T. dubius*, therefore, may have been the

TABLE 37. Development of oat and final densities of nematodes when the natural population of active nematodes was added to heated soil (N/4, N, 4N, 16N). Sowing date 30.7.1969, evaluation date 10.11.1969; cf. Tables 35 and 36 for comparison, and Table 33 for *P.*, *T.*, *R.*, *Tr.*, *S.* and L.S.D.

Treatments	Plant and nematodes	Root weight in g	Shoot weight in g	Shoot length in cm	Final nematode numbers logarithmically transformed <sup>1</sup>					
					in roots		in soil			
					<i>P.</i>		<i>P.</i>	<i>T.</i>	<i>R.</i>	<i>Tr.</i> <i>S.</i>
C (control)		0.97	2.21	50	2.43	2.05	3.26	2.73	2.25	3.85
H (heated, not inoculated)		2.56	4.19	58	0.00	0.00	0.00	0.00	0.00	3.94
N/4		2.14	3.35	61	1.63	0.27	3.20	1.18	0.67	3.88
N		1.80	3.12	61	2.06	0.88	3.51	1.74	1.58	3.78
4 N		1.00	2.29	58	1.99	1.79	3.27	2.31	2.49	3.77
16 N		1.46	2.71	63	2.56	1.56	3.77	2.85	2.87	3.94
L.S.D.		0.40	0.77	5.1	0.335	0.491	0.466	0.418	0.457	N.S.

<sup>1</sup> The initial nematode densities in the control soil C, just before treatments were per tube: *P.* = 2475, *T.* = 1575, *R.* = 735, and *S.* = 6550, or logarithmically transformed 3.39, 3.20, 2.87 and 3.82 respectively.

The density of the extracted and inoculated nematode suspension was for N (number inoculated per tube which was comparable to the normal natural density): *P.* = 2275, *T.* = 1800, *R.* = 865, *Tr.* = 55 and *S.* = 5470, or logarithmically transformed 3.36, 3.26, 2.93, 1.74 and 3.74 respectively.

main parasite. *P. crenatus*, *R. robustus* and even *Tr. pachydermus* can, however, not be ruled out as contributors.

Also it is not sure that the nematodes were the sole cause. The symptoms were not fully reproduced (root and shoot weights were depressed considerably, but shoot length was not), the densities 4N and 16N hardly reached damage rates comparable to untreated, and micro-organisms in or on the nematode bodies may have contributed to the damage, since the inoculated nematodes were not sterilized.

### 8.2.3 Effect of the inoculation procedure on *P. crenatus*, *T. dubius* and *R. robustus*

The inoculation of plant nematodes in steamed soil appeared to result in defective infection and reproduction in several of the experiments recorded in this chapter. A special experiment was made with the population extracted from PD-garden soil comprising predominantly *P. crenatus*, *T. dubius* and *R. robustus*, to study whether inoculation could indeed cause heavy loss of these nematodes, and whether this loss was due to the inoculation procedure in general or specifically to the unsuitability of steamed soil as a habitat for the nematodes.

A virgin sandy polder soil, on which no crops had yet been grown and which contained no plant nematodes at all, was steamed at 100°C for 1 hour at different dates, namely 17 weeks, 6 weeks or 2 days before the soil was inoculated with nematodes, whereas also untreated soil was used. These four soils were filled into plastic tubes containing 105 ml of soil each. Each tube was inoculated with the same dose of a nematode suspension extracted from the naturally infested PD-garden soil. This dose included 124 *P. crenatus*, 350 *T. dubius*, 400 *R. robustus* and 4590 other stylet-bearing and saprozoic nematodes. The inoculation took place on 19.4.1967. Half of the tubes were kept fallow under room conditions; water was added periodically to prevent excessive drying of the soil. The other tubes were sown with ryegrass, *Lolium perenne* L. 'Perma'; water was regularly added to maintain good growth of the grass. The nematode densities were determined at intervals in 5 replicate tubes of the fallow as well as of the grass-grown series. The results are recorded in Tables 38 A and B.

The tables furnish data relevant to our diagnostic study and give some general information as well.

*P. crenatus* collected from soil and re-inoculated, obviously loses the major part of the infective potency of its population. The numerical loss is, on average for all soils, 57% after the 1st week, 88% after 4 weeks, and 96% after 18 weeks. This must be due to kill or inactivation of the nematodes, for losses due to the extraction procedure do not account for more than 20% and do not increase with time. The effects for the untreated and heated soils are similar, and must therefore be due to the inoculation procedure in general, and not to the particular fact that the soils were heated. The inoculated population is thoroughly disorganized, for only after 16 weeks some very weak and erratic multiplication could be observed on ryegrass in one of the soils.

*T. dubius* on average had lost 67% of its population after 1 week, 76% after

TABLE 38. Numbers of *Pratylenchus crenatus* (P.), *Tylenchorhynchus dubius* (T.), *Rorylenchus robustus* (R.), and other stylet-bearing + saprozoic nematodes (O. + S.), at different dates, when the natural mixture of nematodes from PD-garden soil was inoculated into plastic tubes with 105 ml of virgin polder soil, which was either untreated, or heated 2 days, or 6 weeks, or 17 weeks before inoculation. Inoculation date 19.4.1967. Figures are nematode densities in the inoculum added per tube (Inoc.), or extracted from the soil again after one week (+1 week) etc.; all figures are averages of five replicate tubes. Between brackets the logarithmic average values. A = fallow soil, B = soil grown with ryegrass. L.S.D. = least significant difference at 5% level.

Soils		A. Nematodes with time in fallow soils				P.				T.				R.				O. + S.			
		Inoc.	+ 1 w.	+ 4 w.	+ 18 w.	Inoc.	+ 1 w.	+ 4 w.	+ 18 w.	Inoc.	+ 1 w.	+ 4 w.	+ 18 w.	Inoc.	+ 1 w.	+ 4 w.	+ 18 w.	Inoc.	+ 1 w.	+ 4 w.	+ 18 w.
Heated + 2 days		124	52 (1.71)	12 (1.01)	1 (0.18)	350	115 (2.06)	73 (1.85)	23 (1.31)	400	87 (1.92)	75 (1.86)	7 (0.67)	4590	2770 (3.44)	17050 (4.23)	4270 (3.61)				
Heated + 6 weeks		124	43 (1.60)	16 (1.16)	2 (0.31)	350	110 (2.04)	90 (1.95)	53 (1.64)	400	107 (2.02)	110 (1.99)	14 (1.06)	4590	4100 (3.61)	6910 (3.83)	6600 (3.81)				
Heated + 17 weeks		124	34 (1.49)	10 (0.94)	9 (0.84)	350	111 (2.04)	83 (1.89)	69 (1.70)	400	163 (2.21)	156 (2.18)	33 (1.49)	4590	2070 (3.32)	11040 (4.03)	8270 (3.90)				
Untreated		124	58 (1.73)	16 (1.18)	6 (0.62)	350	124 (2.09)	91 (1.94)	26 (1.37)	400	164 (2.21)	107 (2.02)	62 (1.60)	4590	2400 (3.48)	1920 (3.30)	7940 (3.89)				
L.S.D.		-	(N.S.)	(N.S.)	(0.611)	-	(N.S.)	(N.S.)	(N.S.)	-	(0.108)	(0.206)	(0.582)	-	(0.100)	(0.237)	(0.213)				
Soils		B. Nematodes with time in soil grown with ryegrass				P.				T.				R.				O. + S.			
		Inoc.	+ 3 w.	+ 6 w.	+ 16 w.	Inoc.	+ 3 w.	+ 6 w.	+ 16 w.	Inoc.	+ 3 w.	+ 6 w.	+ 16 w.	Inoc.	+ 3 w.	+ 6 w.	+ 16 w.	Inoc.	+ 3 w.	+ 6 w.	+ 16 w.
Heated + 2 days		124	19 (1.18)	3 (0.36)	0 (0.00)	350	86 (1.93)	197 (2.18)	2020 (3.29)	400	89 (1.93)	47 (1.64)	135 (2.13)	4590	3510 (3.52)	12570 (4.09)	4730 (3.65)				
Heated + 6 weeks		124	12 (1.05)	5 (0.57)	1 (0.16)	350	71 (1.85)	776 (2.87)	2110 (3.32)	400	95 (1.97)	156 (2.17)	141 (2.12)	4590	11350 (4.05)	8870 (3.94)	5090 (3.70)				
Heated + 17 weeks		124	10 (0.94)	5 (0.52)	14 (1.12)	350	114 (2.04)	878 (2.93)	1610 (3.19)	400	148 (2.15)	136 (2.12)	303 (2.45)	4590	6100 (3.78)	10250 (3.99)	3400 (3.51)				
Untreated		124	9 (0.76)	2 (0.31)	2 (0.21)	350	93 (1.96)	225 (2.26)	810 (2.85)	400	146 (2.16)	73 (1.85)	133 (2.01)	4590	1740 (3.24)	1380 (3.14)	3100 (3.44)				
L.S.D.		-	(N.S.)	(N.S.)	(0.438)	-	(N.S.)	(0.323)	(0.228)	-	(0.187)	(0.193)	(N.S.)	-	(0.159)	(0.157)	(N.S.)				

4 weeks and 88% after 18 weeks. Untreated soil was not different from the heated soils in the fallow series. In the grass-grown series, however, significant differences between the soils occurred for nematode reproduction, which generally showed up after 6 weeks and was strong after 16 weeks. Reproduction in the untreated soil was less than in the heated soils after 6 and after 16 weeks. A further check, which is not recorded in the table, confirmed that after 24 weeks this difference was still present and, therefore, must be considered a long-term effect. In the soils heated only 2 days before inoculation the reproduction was delayed after 6 weeks, but no longer after 16 weeks.

*R. robustus* had on average for all soils lost 67% of its population after 1 week, 72% after 4 weeks and 93% after 18 weeks. For this nematode, however, the loss was higher if the period between heating and inoculation had been shorter. After 1, 4, and 18 weeks 22%, 19% and 2% of the population was recovered from the 'H + 2 days' soil; 41%, 38% and 8% from the 'H + 17 weeks' soil; and 41%, 27% and 16% from the untreated fallow soil. In the grass-grown series the same difference between soils was visible after 3 weeks, and also after 6 weeks although at that moment nematode reproduction had started to show effect. After 16 weeks the 'H + 2 days' soil still caused delay of reproduction as compared to 'H + 6 weeks' and 'H + 17 weeks', but at this moment untreated soil also fell behind 'H + 17 weeks'. An extra check after 24 weeks demonstrated that this difference was maintained, as for *T. dubius*.

The saprozoic nematodes demonstrated some marked phenomena. In fallow soil their numbers were somewhat below the inoculated dose for all soils after 1 week, but then strong temporary reproduction occurred in all heated soils, but not in the untreated soil. After 18 weeks this marked density peak had finished. The same phenomenon was visible in the grass-grown soils. Heated soil apparently is a very suitable substrate for saprozoic nematodes for a period which is more than 6 but shorter than 16 weeks. This may be due to selective reproduction of the inoculated nematodes on the decaying remains of the killed soil fauna and flora. After 16 to 18 weeks the numbers are again the same as for untreated soil. This temporary gradation of saprozoic nematodes may influence the establishment and reproduction of plant parasitic species in the same soil.

The results fit in with the experience gained in the inoculation experiments described under 8.2.1. and 8.2.2. and explain why the effects of inoculation were often erratic and obscure.

### 8.3. DISCUSSION

The experiments show on the one hand that inoculation with the plant nematodes reproduced the oat yield deficiency to a large extent. On the other hand they demonstrate special difficulties encountered in inoculation experiments with nematodes.



The presence of oat yield deficiency in the PD-garden soil was clearly demonstrated by PSS in all experiments. Inoculation of uncleaned soil-extract to heated soil fully reproduced the disease syndrome (Tables 33 and 35, Figs. 26 and 27), unless the extract was strongly blended and dried so that the nematodes were killed (Table 36). Inoculation of the nematodes, i.e. the whole natural population of actively moving specimens, was effective (Table 36), although the introductory experiment of the same set-up had been inconclusive, possibly because the nematodes established themselves too poorly (Table 33). Inoculation of *P. crenatus* on infected rootlets did not lead to a high population density and did hardly or not at all depress plant growth. Inoculations of suspensions of nematodes were complicated by defective initial infection, by decline of the populations when the host plant was heavily damaged, and in some of the experiments also by decline of the natural population in the untreated control soil. The defective initial infection must be due to the fact that many nematodes could not endure the inoculation procedure, at least not as suspensions in water, according to the special experiments, about the fate of our experimental nematodes upon inoculation into soil. The results of Table 38 namely show, that 70–90% of inoculated *P. crenatus*, *T. dubius* and *R. robustus* are lost within a few weeks after inoculation into soil, even when a host plant is grown. The actual infective potential, therefore, is only a fraction of the densities inoculated into soil. This holds for untreated as well as for heated soil, although heating may enhance the effect for some species. As soon as the initial unfavourable effect of heating has disappeared, heated soil appears to allow more rapid reproduction of *T. dubius* and *R. robustus* than untreated soil, due to the freedom from specific enemies, diseases and predators, existing in natural niches, in the new loci. *P. crenatus*, however, was influenced by the inoculation to such an extent that its population nearly collapsed and did not regain noticeable reproduction for several months. Defective infection due to the inoculation procedure must have influenced all our inoculation experiments, and it may explain the difficulty to obtain conclusive data on the pathogenicity of nematodes in general.

The results indicate that the oat problem studied is at any rate associated with nematode infestation, although the role played by each of the prevailing species is not determined and the contribution by microorganisms is not excluded (cf. Fig. 26). In inoculation experiments with nematodes heavy losses due to manipulation of the nematodes should be taken into account: initial densities cannot be derived from the inoculation doses alone, but must be assessed by extraction after the inoculation to determine the survivors.

## 9. SUMMARY

Research was carried out on the possible yield increase of crops in The Netherlands by the use of PSS (partial soil sterilization) on the soil, on the basis of published as well as unpublished data and by experimentation with different disinfectants, soils and plants.

Following review of the literature and the methods employed are chapters dealing with inventory of quantitative data, PSS treatments of random fields without known problems, of soils with known nematode problems, and of fresh uninfested soils, with a final chapter on diagnostic studies on one particular PSS effect in oat.

PSS was applied to 600,000 ha, 1 % of the land, in the U.S.A. and to 40,000 ha, 4 % of the arable land, in The Netherlands in 1969. The literature of the last five years comprises more than 400 scientific articles, but no reliable estimates have apparently been published about the magnitude of the effect of PSS in general, nor has the effect on plant growth been fully explained. The effect of the various PSS treatments, physical as well as chemical, may be direct in connection with the ingredients applied or the soil organisms or the soil itself, and indirect, e.g. in connection with plant growth. Plant growth is the main parameter used to measure PSS effects in this study, although density of pathogens (especially nematodes), and the chemical soilfertility (especially in connection with nitrogen) have sometimes been assessed as well.

The approach and the methods employed in this study were partly statistical and partly experimental; in both cases common techniques were used. The data were usually arranged after calculations and tested by means of the IBM and CDC computerfacilities of the Landbouwhogeschool.

Altogether 2453 individual treatment-soil-plant combinations from trials in The Netherlands during the period 1947-1970 could be collected. They were derived from the author's 545 experiments, from unpublished files of the Plantenziektenkundige Dienst, the Landbouwhogeschool and other institutions or individual researchers, and from literature. For each separate item the growth or yield was calculated as a percentage of untreated, and inclusive of supplementary data inserted into a computer card. After standardization and comprehension of the data special computer programmes furnished a number of tables and diagrams (Tables 2-9, Figs. 1-6). The yield on treated soil showed as general average 141 % of untreated, or 154 % without phytotoxic cases. For PSS treatments with the best known chemicals these figures were 151 %/164 %. If for each crop the average best treatment is selected, 172 %/188 % is reached. The average results of the treatments increased from pentachloronitrobenzene (103 %/118 %), via Vapam, Trapex, dichloropropene and chloropicrin to heat (221 %/240 %). The recommended doses normally used in practice appeared to give optimal average PSS effects. The percentage phytotoxic cases was on an average about 20 %. It did not increase with higher doses, but was strongly

correlated with the boiling point of the fumigants used. The crop averages differed widely. Pea, barley and woody plants seldom reached a good yield unless PSS had been applied. For grass, clover, potato and cabbage an intermediate yield increase was usually reached, whereas the response to PSS of many crops was low on some and very high on other soils. The measure of growth improvement was strongly correlated to the presence of known noxious pests and diseases (Tables 5-7). They were according to this inventory, very low on new polder soil, or clean sand, or fresh peat (99%/114%), and moderate on random soils without known infestation (115%/125%), as compared to the overall average already mentioned above (141%/154%). The effects varied much with soil type; they decreased from loamy sand, via clay soil, peaty sand, diluvial sand and dune sand to silty clay. The effect for polder sand (mainly fresh soils) was slight and for river sand it was even negligibly small, indicating the absence of stimulatory effects on plant growth by the chemicals themselves. PSS effects also varied with the different localities throughout the country. The relatively high effects for the Winschoten and Wageningen districts are the result of numerous experiments on heavily nematode-infested plots and experiments with imported loads of infested test soils. Therefore they are not representative for these districts. The low effect for Emmeloord district reflects results obtained on new, uninfested polder soils.

The PSS treatments on 11 random fields without known problems (treatments with chloropicrin, dichloropropene and pentachloronitrobenzene, 7 to 14 test crops per field, three replicates) reached an average yield of 113%, or 122% excluding the phytotoxic cases, or 156% for the average of the best treatments of each soil-crop combination. Nearly all crops suffered heavy yield losses on some fields and nearly all fields contained noxious problems for some crops which could be solved by PSS treatment (Fig. 7). The potential effect of PSS applied to all agricultural land including the problem fields must exceed 156%, but should be less than 188% found above as the average for all trials in The Netherlands.

Five PSS treatments applied in pot trials to eight soils, with a known, noxious nematode problem and grown with nematode susceptible test plants, illustrated strong growth responses according to expectation, except for two soils where nematodes were probably not the main nor the sole problem, and except for some phytotoxic effects (Tables 11-26, Figs. 9-19). Dichloropropene was as effective as heat and chloropicrin in these soils with respect to nematode control and reached about the same yield increases. The nematicidal effect of Temik was delayed but sustained; its systemic potencies were confirmed, but sometimes it was phytotoxic. Captan showed poor nematicidal, but not wholly negligible effect. It was among the best treatments only in one soil with a suspected fungus infestation (Fig. 19) and was strongly phytotoxic in two soils (Figs. 17, 18).

In pot trials with five PSS treatments of soils without or with very few noxious organisms caused only barely measurable growth responses to the crops carrot, rose and ryegrass (Tables 29, 31 and Figs. 20-24). Two uniden-

tified, probably seed-borne problems, i.e. loss of seedlings and branching of roots, encountered in carrot, did not show up in soils previously disinfected with heat, chloropicrin or a very heavy dose of dichloropropene (Fig. 21).

Diagnostic studies on an important PSS effect on oat, in soils infested with a complex of freeliving root-infesting nematodes (*Pratylenchus crenatus* + *Tylenchorhynchus dubius* + *Rotylenchus robustus*) proved, that inoculation with the nematode could produce the damage for the greater part (Tables 33–37, Figs. 25–27). The possible contribution of other organisms was however not excluded. The inoculation procedure seemed to cause important losses amongst the nematodes thus inoculated. For some species these were even aggravated by using steamed or heated soil shortly prior to the test.

## 10. SAMENVATTING

Onderzoek werd verricht over de mogelijke opbrengstvermeerdering van gewassen in Nederland door partiële sterilisatie (PS) van de grond, aan de hand van gepubliceerde en ongepubliceerde gegevens en proeven met verschillende ontsmettingsmiddelen, gronden en planten.

Na overzichten van de literatuur en van de gebruikte methoden, volgen hoofdstukken over de inventarisatie van quantitative gegevens, PS behandelingen van willekeurige velden zonder bekende problemen, van gronden met bekende aaltjesproblemen en van nieuwe of onbesmette gronden en een afsluitend hoofdstuk over diagnostisch onderzoek van een speciaal PS-effect in haver.

In 1969 werd PS toegepast op 600.000 ha, 1 % van de grond, in de USA en op 40.000 ha, 4 % van het bouwland, in Nederland. De literatuur van de laatste 5 jaren omvat meer dan 400 wetenschappelijke artikelen, maar betrouwbare gegevens over de grootte van het PS-effect in het algemeen zijn blijkbaar niet bekend en ook is het effect van PS op de plantengroei niet volledig verklaard. De invloed van verschillende PS-behandelingen, fysische en chemische, kan direct zijn, verband houdende met de toegediende stof of de bodem-organismen of de grond zelf en indirect, bijvoorbeeld met betrekking tot de groei van de plant. Groei van de plant is in deze studie de voornaamste parameter voor het meten van PS-effecten, hoewel de besmettingsgraad met pathogenen (in het bijzonder nematoden) en de chemische vruchtbaarheidstoestand van de grond (vooral met betrekking tot stikstof) soms ook zijn bepaald en gebruikt.

De benadering en de gebruikte methoden zijn ten dele statistisch en ten dele experimenteel; in beide gevallen werden bekende technieken gebruikt. De gegevens werden meestal gerangschikt, berekend en getoetst met behulp van de IBM en CDC computer-faciliteiten van de Landbouwhogeschool.

In totaal konden 2453 behandeling-grond-plant combinaties van in de jaren 1947-'70 in Nederland verrichtte proeven bijeengebracht worden. Zij waren afkomstig (545) van de schrijver, van ongepubliceerde proeven van de Plantenziektenkundige Dienst, de Landbouwhogeschool, andere instellingen en individuele onderzoekers en uit de literatuur. Van elke behandeling afzonderlijk werd de groei of opbrengst van het gewas berekend als een percentage van onbehandeld en met bijbehorende gegevens op een computerkaart gebracht. Na standaardisatie en comprimering van de gegevens verschaften speciale computer-programma's een aantal overzichtstabellen en diagrammen (Tab. 1-9, Fig. 1-6).

De opbrengst van behandelde grond was als algemeen gemiddelde 141 % van onbehandeld of 154 % zonder de gevallen met phytotoxiciteit. Voor behandelingen met de gangbare grondontsmettingsmiddelen waren deze cijfers 151 %/164 %. Als voor elk gewas de gemiddeld beste behandeling wordt gekozen, werd 172 %/188 % bereikt. De gemiddelde resultaten voor de middelen stegen van

pentachloornitrobenzeen (103%/118%) via Vapam, Trapex, dichloorpropeen, en chloorpicrine naar stomen van de grond (221%/240%). De voor de praktijk als normaal aanbevolen doseringen bleken gemiddeld optimale resultaten te geven. Het percentage gevallen met phytotoxiciteit was gemiddeld ongeveer 20; dit nam niet toe bij de hogere doseringen in de proeven maar bleek sterk gecorreleerd te zijn met het kookpunt van de gebruikte fumigantia.

De gewas-gemiddelden varieerden sterk. Erwt, gerst en houtige gewassen bereikten zelden een goede opbrengst tenzij PS werd toegepast. Voor gras, klaver, aardappel en kool werd meestal een middelmatige opbrengstvermeerdering bereikt, terwijl de groei van vele gewassen op sommige gronden weinig en op andere zeer sterk werd vermeerderd door PS. De mate van groeiverbetering was sterk gecorreleerd met de aanwezigheid van bekende, schadelijke ziekten en plagen (Tab. 5-7). Ze was volgens deze inventarisatie zeer gering op nieuwe poldergrond of rivierzand of verse turfgrond (99%/114%) en matig op willekeurig gekozen gronden zonder bekende besmetting (115%/125%), vergeleken met het reeds genoemde algemene gemiddelde (141%/154%). Zij varieerde sterk met de grondsoort en daalde in de volgorde lemige zandgrond, kleigrond, dalgrond, diluviale zandgrond, duinzandgrond, zavel. Op polderzandgrond, voornamelijk nieuwe grond, was het effect gering en in rivierzandgrond zelfs verwaarloosbaar klein, hetgeen er op wijst dat de chemicaliën zelf de plantengroei niet verbeterden. Het effect was ook verschillend voor verschillende districten van het land. De relatief sterke effecten in de districten Winschoten en Wageningen zijn een gevolg van veel proeven op zwaar met aaltjes besmette percelen voor deze districten. Het lage effect in het district Emmeloord betreft weer vooral het resultaat op nieuwe, onbesmette poldergrond.

De speciale proeven met PS op 11 willekeurig gekozen velden zonder bekende problemen (behandelingen met chloorpicrine, dichloorpropeen en pentachloornitrobenzeen, 7-14 gewassen per veld, 3 herhalingen) leverden een gemiddeld opbrengstcijfer van 113% of 122% zonder de phytotoxische gevallen of 156% voor het gemiddelde van de beste behandelingen van elke grond-gewas combinatie. Bijna alle gewassen leden ernstige opbrengstverliezen op enkele velden en bijna alle velden bevatten door PS te verhelpen problemen voor enkele gewassen (Fig. 7.). Het potentiële effect van PS op alle cultuurgrond inclusief de probleemvelden toegepast, moet meer zijn dan 156% maar is minder dan 188%, het eerder genoemde gemiddelde van de beste gewas-behandelingen gebaseerd op alle proeven in Nederland.

Vijf PS-behandelingen werden in potproeven toegepast op acht gronden met een bekend schadelijk aaltjesprobleem; daarna werden voor de aaltjes gevoelige toetsgewassen geteeld. Zij veroorzaakten grote opbrengstvermeerderingen, volgens de verwachting, behalve op twee gronden waar aaltjes waarschijnlijk niet het voornaamste of niet het enige probleem vormden en afgezien van enkele phytotoxische effecten (Tab. 11-26, Fig. 9-19). Dichloorpropeen was in deze gronden tegen aaltjes even effectief als stomen of chloorpicrine en bereikte ongeveer dezelfde opbrengstvermeerderingen. Temik had op de aaltjes een

langzaam-dodend maar lang aanhoudend effect; de systemische werking van dit middel werd bevestigd, maar het bleek soms phytotoxisch te zijn. Captan gaf een slecht, maar niet geheel te verwaarlozen resultaat tegen de aaltjes. Het middel behoorde alleen tot de beste in een grond waarin een schimmelaantasting waarschijnlijk van belang was (Fig. 19) en het was sterk phytotoxisch in twee gronden (Fig. 17, 18).

In potproeven met vijf PS-behandelingen van gronden zonder of vrijwel zonder schadelijke organismen werden nauwelijks meetbare groeiverbeteringen bij peen, roos en Engels raaigras verkregen (Tab. 29–31, Fig. 20–24). Twee niet-geïdentificeerde, mogelijk door het zaad ingebrachte problemen bij peen, namelijk verlies van zaailingen en wortelvertakking, waren niet merkbaar in gronden die tevoren waren ontsmet met hitte, chloorpicrine of een zeer zware dosis dichloorpropeen (Fig. 21).

Diagnostisch onderzoek over een belangrijk PS-effect in haver op gronden met een complex van vrijlevende wortelaaltjes (*Pratylenchus crenatus* + *Tylenchorhynchus dubius* + *Rotylenchus robustus*) toonde aan, dat inoculatie met de aaltjes de schade grotendeels kon reproduceren (Tab. 33–37, Fig. 25–27); de mogelijke bijdrage van andere organismen werd evenwel niet uitgesloten. De inoculatie-procedure bleek belangrijke verliezen onder de geïnoculeerde aaltjes te veroorzaken, die voor sommige soorten nog versterkt werden bij het gebruik van kort voor de proef gestoomde grond.

## II. ACKNOWLEDGEMENTS

To Dr. Ir. M. Oostenbrink, I express my sincere gratitude and indebtedness for suggesting the research topic, his guidance and supervision, furnishing working facilities in his Departments at the Plantenziektenkundige Dienst (PD) and the Landbouwhogeschool (LH) as well as the financial support and for his valuable constructive criticism and support in preparing the manuscript. I feel sorry that he cannot be present at the promotion ceremony due to his stay in India. I am very much indebted to Dr. Ir. J. Dekker, professor of phytopathology as well for his interest in the subject, his cooperation and sincere willingness to act as promotor.

The efforts and help of my Dutch colleagues at the PD and the LH, I greatly appreciate. I also extend my sincere gratitude to Ir. M. J. Hijink, Ir. K. Kuiper, Mr. S. Stermerding Ing., Dr. Ir. H. Hoestra, Ir. J. A. van Berkum, Mr. H. Hietbrink Ing., Ir. W. P. de Leeuw, Drs. P. A. A. Loof, Drs. G. H. Boerema and Miss H. Sanders and their coworkers who effectively helped and advised me in realizing my research scheme. Mr. E. Immikhuizen as well as Dr. Ir. H. Hoestra supported my work by providing unpublished data.

Special thanks are due to Mr. D. Dijkstra Ing., Mr. M. Keuls, Ir. J. Spiertz and Mr. C. A. Hoveyn for their inestimable cooperation in the statistical approach, teaching me to handle modern computing systems and using the implements concerned.

I am most grateful to Mr. R. Havlik who did most of the drawings and Mr. D. Lindner who photographed them. Also to Mr. J. P. Jannette-Walen Ing. for rectifying the English text, as well as Mrs. M. E. Brink-de Boois, Miss M. A. Peters, Mrs. H. I. M. Wilthagen for their help in typing the manuscript and Mr. H. Viets for revising the literature list.

I finally wish to express my gratitude to my colleagues in Egypt who contributed to this work, to the Egyptian National Research Centre in Cairo and the U.A.R. Embassy staff members at The Hague for their permission and support to take leave and realize this study.



## 12. REFERENCES

- ADAMS, R. E. & TRUE, R. P. (1962). The response of Norway spruce to soil fumigation for the control of nematodes. *Phytopathology* 52: 1216.
- ALLEN, M. W. & RASKI, D. J. (1952). Soil fumigation to control root-lesion nematode, *Pratylenchus* sp., in tuberous begonia. *Pl. Dis. Repr* 36: 201-202.
- ALLEN, M. W. & MAGGENTI, A. R. (1959). Plant nematology in California. *Calif. Agric.* 13(9): 2-3.
- ALTMAN, J. (1963). Increase in nitrogenous compounds in soil following soil fumigation. *Phytopathology* 53: 870.
- ALTMAN, J. & LAWLOR S. (1964). The effect of chlorinated C<sub>3</sub> hydrocarbons on growth and amino acid production of indigenous soil bacteria. *Phytopathology* 54: 886.
- ANDERSON, E. J. (1956). Comparison of initial kills and subsequent increase of nematode populations following soil fumigation. *Phytopathology* 46: 634.
- ANDERSON, P. J. (1951). Control of tobacco nematodes by soil fumigation. *Phytopathology* 41: 657.
- ANONYMOUS (1950a). Control of Fusarium root rot of peas with fumigants. *Pl. Dis. Repr* Suppl. 192: 179.
- ANONYMOUS (1950b). Results with soil fumigation and drench tests. *Pl. Dis. Repr* Suppl. 192: 178-179.
- ANONYMOUS (1968). Principles of plant and animal pest control. Volume 4. Control of plant-parasitic nematodes. *Publ. Natn. Acad. Sci., Wash.* 1696: 172 p.
- APT, W. J. & GOULD, C. J. (1961). Control of root-lesion nematode, *Pratylenchus penetrans* on narcissus. *Pl. Dis. Repr* 45: 290-295.
- ASHWORTH, L. J., LANGLEY, B. C. & THAMES, W. H. (1964). Long-term inhibition of *Rhizoctonia solani* by a nematocide, 1,2-dibromo-3-chloropropane. *Phytopathology* 54: 187-191.
- ATKINS, J. G. & FIELDING, M. J. (1956). A preliminary report on the response of rice to soil fumigation for the control of stylet nematodes, *Tylenchorhynchus martini*. *Pl. Dis. Repr* 40: 488-489.
- BAERMANN, G. (1917). Eine einfache Methode zur Auffindung von Ankylostomum-Larven in Erdproben. *Geneesk. Tijdschr. Ned. Indië* 57: 131-137.
- BAINES, R. C. & MARTIN, J. P. (1953). Effect of soil fumigation on growth and yield of Valencia orange trees. *Phytopathology* 43: 465-466.
- , SMALL, R. H., DE WOLFE, T. A., MARTIN, J. P. & STOLZY, L. H. (1957). Control of the citrus nematode and *Phytophthora* spp. by Vapam. *Pl. Dis. Repr* 41: 405-414.
- , DE WOLFE, T. A. & SMALL, R. H. (1958). Control of the citrus nematode, *Phytophthora* spp. and weeds by Mylone 85 W when applied by different methods. *Pl. Dis. Repr* 42: 876-880.
- , MARTIN, J. P., DE WOLFE, T. A., BOSWELL, S. B. & GARBER, M. J. (1962). Effect of high doses of D D on soil organisms and the growth and yield of lemon trees. *Phytopathology* 52: 723.
- BAKER, A. D., OOSTENBRINK, M. & BERKUM, J. A. VAN (1967). Bibliography on plant, soil and freshwater nematodes (with supplements). BRILL, Leiden.
- BAKER, K. F. & OLSEN, C. M. (1960). Aerated steam for soil treatment. *Phytopathology* 50: 82.
- BAKKER, H. DE & SCHELLING, J. (1966). Systeem van bodemclassificatie voor Nederland. PUDOC, Wageningen 1966.
- BARKER, K. R. & SASSER, J. N. (1959). Biology and control of the stem nematode, *Ditylenchus dipsaci*. *Phytopathology* 49: 664-670.
- BELEHRADEK, J. (1935). Temperature and living matter. *Protoplasma-Monogr.* 8: 277 p.
- BERGESON, G. B. (1955). The use of systemic phosphates for control of *D. dipsaci* on alfalfa and daffodils. *Pl. Dis. Repr* 39: 705-709.

- BERKUM, J. A. VAN (1964). Nematocidal value of some simple organic compounds. *Nematologica* 10: 62.
- BESEMER, A. F. H. & OOSTENBRINK, M. (1955). Phytotoxische en nematicide nawerking van grondontsmettingen met DD. Meded. LandbHoogesch. OpzoekStns Gent 20: 279-290.
- BIRCHFIELD, W. & PINCKARD, J. A. (1964). New combinations of nematocides for control of reniform nematode on cotton. *Phytopathology* 54: 393-394.
- BIRD, G. W. & SMITH, H. A. (1965). Factors affecting control of onion bloat by fumigants containing 1,3-dichloropropene in organic soils in southern New York. *Pl. Dis. Repr* 49: 33.
- BIRD, L. S. (1963). In-covering soil fungicides, nematocides and insecticides for disease control in cotton. *Phytopathology* 53: 622.
- BLOOM, J. R. (1963). Lethal effect of temperature extremes on *Anguina tritici*. *Phytopathology* 53: 347.
- BOOGAART, K. VAN DEN & HUIJK, M. J. (1959). Tridipam, een nieuw nematicide. Meded. LandbHoogesch. OpzoekStns Gent 24: 645-661.
- BRANDE, J. VAN DEN & GILLARD, A. (1957). Bestrijding van wortelknobbelaaltjes (*Meloidogyne* spp.) door elektrische bodemverwarming. *Tuinbouwberichten* 21: 63-64.
- BROWN, E. B. & FRANKLIN, M. T. (1953). Experiments on control of eelworm in black currants. *Pl. Path.* 2: 101-102.
- BROWN, K. F. (1963). Nematodes, destroyers of the world's crops. *Span* 6: 98-100.
- BROWN, K. F. & DUNN, C. L. (1965). The uptake of carbon-14 labelled ethylene dibromide nematicide by *Ditylenchus dipsaci* (KÜHN). *Nematologica* 11: 353-360.
- BROWN, L. N. (1934). Flooding to control root-knot nematodes. *J. agric. Res.* 47: 883-888.
- BUCHENAU, G. W. (1963). Winter survival of wheat in chloropicrin treated soil. *Pl. Dis. Repr* 47: 602-604.
- BYARS, L. P. (1919). Experiments on the control of the root-knot nematode, *Heterodera radicola* (GREEF) MUELLER. 1. The use of hydrocyanic-acid gas in loam soil in the field. *Phytopathology* 9: 93-104.
- CAIRNS, E. J. (1955). Nematodes, tiny but mighty. Research under way points to development of better and cheaper controls. *Highlts agric. Res.* 2 (1): 4-5.
- CALVERT, E. L. & HARRISON, B. D. (1966). Potato mop-top, a soil-borne virus. *Pl. Path.* 15: 134-139.
- CARTER, W. (1943). A promising new soil amendment and disinfectant. *Science* 97: 383-384.
- ČEKALINA, V. I. (1966). Chimičeskie preparaty dlja bor'by s nematodami. 3. Ditiokarbarnaty, tiocianaty, geterocikličeskie soedinenija, polisulfidy i drugie proizvodnye sery. *Chimija v sel'skom chozjajstve*, Nr. 3: 35-45.
- CHAMBERS, A. Y. & REED, H. E. (1963). Effectiveness of application methods with several nematocides for control of root-knot nematode. *Phytopathology* 53: 622.
- CHANDRA, P. & BOLLEN, W. B. (1961). Effects of Nabam and Mylone on nitrification, soil respiration and microbial numbers in four Oregon soils. *Soil Sci.* 92: 387-393.
- CHEN, T. A., KILPATRICK, R. A. & RICH, A. E. (1962). Influence of dieldrin and methyl bromide soil treatments on subsequent populations of stylet-bearing nematodes associated with roots of Ladino clover. *Phytopathology* 52: 922.
- CHEVRES-ROMAN, R. (1967). Influence of certain nematodes and number of consecutive plantings of sorghum and corn on forage production, chemical composition of plant and soil, and water use efficiency. *Nematologica* 13: 139.
- CHITWOOD, B. G. (1941). Soil treatments with volatile liquids for control of nematodes. *Phytopathology* 31: 818-824.
- & NEWHALL, A. G. (1942). The efficacy of certain nematocides in the control of onion bloat in muck soil. *Phytopathology* 32: 254-258.
- (1952). Nematocidal action of halogenated hydrocarbons. *Adv. Chem. Ser.* 7: 91-99.
- CHRISTIE, J. R. (1945). Some preliminary tests to determine the efficacy of certain substances when used as soil fumigants to control the root-knot nematode, *Heterodera marioni* (CORNU) GOODEY. *Proc. helminth. Soc. Wash.* 12: 14-19.
- (1947a). Preliminary tests to determine the nematocidal and fungicidal properties of certain chemical compounds when used as soil fumigants. *Proc. helminth. Soc. Wash.* 14: 23-28.

- CHRISTIE, J. R. (1947b). Soil fumigation for control of nematodes and other soil-inhabiting organisms. *Pl. Dis. Reprtr Suppl.* 170: 169-189.
- (1959). Plant nematodes, their bionomics and control. *Agric. Exp. Stns Univ. Fla., Gainesville*: 256 p.
- COLBRAN, R. C. and GREEN, H. T. (1963). Effect of time of application of nematocide on the halide content of flue-cured tobacco leaf. *Qd J. agric. Sci.* 20: 101-107.
- CONVERSE, R. H. (1960). Control of *Phytophthora fragariae* with soil fungicides. *Pl. Dis. Reprtr* 44: 948-951.
- COOPER, W. E., WELLS, J. C. & SASSER, J. N. (1959). Sting nematode control on four crops with pre- and post plant applications of Nemagon. *Phytopathology* 49: 316.
- CRALLEY, E. M. (1957). The effect of seedling methods on the severity of white tip of rice. *Phytopathology* 47: 7.
- CRITTENDEN, H. W. (1953). Effect of clean fallow in root-knot development on soybeans. *Phytopathology* 43: 405.
- DAO, F. (1970). Climatic influence on the distribution pattern of plant parasitic and soil inhabiting nematodes. *Meded. Landbouhogesch. Wageningen* 70 (2): 181 p.
- DARBY, J. F. (1961). Evaluation of treatments for the control of soil-borne pests of tomato. *Pl. Dis. Reprtr* 45: 58-61.
- , DIETER, C. E. & RAU, G. J. (1962). Evaluation of treatments for control of soil-borne pests in celery seedbeds. *Pl. Dis. Reprtr* 46: 441-443.
- DARLING, H. M. (1959). Control of the potato root nematode in Wisconsin. *Pl. Dis. Reprtr* 43: 239-242.
- DAULTON, R. A. C. & STOKES, W. M. (1952). The destruction or inhibition of root-knot nematodes by exposure to an electrostatic field. *Emp. J. exp. Agric.* 20: 271-273.
- DECKER, H. (1963). Pflanzenparasitäre Nematoden und ihre Bekämpfung. VEB dt. Landwirtschaftsverlag, Berl.: 374 p.
- DEVAY, J. E., LUKEZIC, F. L. & ENGLISH, W. H. (1962). Effect of DD and various dichloropropenes on the resistance of peach seedlings to bacterial canker. *Phytopathology* 52: 729.
- DICKEY, R. S. (1962). Efficacy of five fumigants for the control of *Agrobacterium tumefaciens* at various depths in the soil. *Pl. Dis. Reprtr* 46: 73-76.
- DIMOCK, A. W. (1944). Soil treatment with sodium selenate for control of foliar nematode of chrysanthemums. *Phytopathology* 34: 999.
- & FORD, C. H. (1950). Control of foliar nematode disease of chrysanthemums with parathion sprays. *Phytopathology* 40: 7.
- DOMSCH, K. H. (1959). Untersuchungen zur Wirkung einiger Bodenfungizide. *Mitt. biol. BundAnst. Ld-u. Forstw.* 97: 100-106.
- DUBUISSON, J. P. (1917). The extraction and saturation of soils with volatile antiseptics. *Soil Sci.* 3: 353-391.
- EDMUNDS, J. E. & MAI, W. F. (1966). Effect of *Trichoderma viride*, *Fusarium oxysporum*, and fungal enzymes upon the penetration of alfalfa roots by *Pratylenchus penetrans*. *Phytopathology* 56: 1132-1135.
- , BOOTHROYD, C. W. & MAI, W. F. (1967). Soil fumigation with DD for control of *Pratylenchus penetrans* in corn. *Pl. Dis. Reprtr* 51: 15-19.
- EISSA, M. F. M. (1971). The basic list for the effect of partial soil sterilization on plant parasitic nematodes and plant growth. Nematology Dept. Plziektenk. Dienst Wageningen (unpublished).
- ENNIK, G. C., KORT, J. & LUESINK, B. (1964). The influence of soil disinfection with DD, certain components of DD and some other compounds with nematocidal activity on the growth of white clover. *Neth. J. Pl. Path.* 70: 117-135.
- FEDER, W. A. (1952). Systox for nematode in daffodils. *Bull. N. Y. Flower Growers Inc.* 82: 3-4.
- (1954). Laboratory evaluation of certain nematicidal materials. *Phytopathology* 44: 428-431.
- , EICHORN, J. L. & HUTCHINS, P. C. (1962). Sugar-induced osmotic dehydration of nematodes enhanced by the addition of detergents. *Phytopathology* 52: 9.

- FECKES, W. (1967). Phytopathological consequences of changing agricultural methods. 2. Cereals. *Neth. J. Pl. Path.* 73, Suppl.: 97-115.
- FINK, H. C. (1956). Vapam and PCNB soil treatments for potato scab control. *Pl. Dis. Repr* 40: 190-192.
- GARRETT, S. D. (1938). Soil conditions and the root-infecting fungi. *Biol. Rev.* 13: 159-185.
- (1956). Biology of root infecting fungi (2. Edn). Camb. Univ. Press, Lond.: 92 p.
- (1964). Soil microbiology for root disease investigators. A course of lectures given at the National Research Centre, Cairo Jan.-April: 145 p.
- GIESE, A. C. (1957). Temperature and life. *Cell Physiology Illustrated*, W. B. Saunders Co., Philadelphia: 52-56.
- GODFREY, G. H. (1934a). The confinement of chloropicrin and other gases for fumigation purposes. *Phytopathology* 24: 1366-1373.
- (1934b). Control of soil fungi with chloropicrin. *Phytopathology* 24: 1146.
- GOOD, J. M. (1964). Effect of soil application and sealing methods on the efficacy of row applications of several soil nematocides for controlling root-knot nematodes, weeds, and Fusarium wilt. *Pl. Dis. Repr* 48: 199-203.
- & RANKIN, H. W. (1964). Evaluation of soil fumigants for control of nematodes, weeds and soil fungi. *Pl. Dis. Repr* 48: 194-199.
- & CARTER, R. L. (1965). Nitrification lag following soil fumigation. *Phytopathology* 55: 1147-1150.
- & FELDMESSER, J. (1966). The future role of chemicals for controlling plant nematodes. In: Pest control by chemical, biological, genetic and physical means, Symp. A. R. S. USDA: 52-57.
- (1968). Relation of plant parasitic nematodes to soil management practices. In: Tropical Nematology, G. C. Smart and V. G. Perry, Univ. Fla Press, Gainesville: 113-138.
- GORING, C. A. I. (1962). Control of nitrification by 2-chloro-6- (trichloromethyl) pyridine. *Soil Sci.* 93: 211-218.
- GRAINGER, J. (1956). Progress in soil mixing for nematode control. *Nematologica* 1: 13-46.
- (1960). Safety in soil treatments for eelworm control. *Nematologica Suppl.* 2: 6-12.
- & CLARK, M. R. M. (1963). Interactions of *Rhizoctonia* and potato root eelworm. *Eur. Potato J.* 6: 131-132.
- GREEN, C. D. & WEBSTER, J. M. (1966). The effect of ultraviolet radiation on the stem and bulb nematode *D. dipsaci* (KÜHN). *Nematologica* 11: 638-642.
- HAGUE, N. G. M. & SOOD, U. (1963). Soil sterilization with methyl bromide to control soil nematodes. *Pl. Path.* 12: 88-90.
- HAMMER, O. H. (1943). Method for controlling parasites in the soil. U. S. Patent No. (1946) 2: 403-435.
- HIJINK, M. J. (1969). Werking en nevenwerkingen van grondontsmettingsmiddelen. *Landbouwk. Tijdschr.*, 's-Grav. 81: 86-92.
- HOESTRA, H. (1961). The role of nematodes in the orchard replant problem. 6th Int. Nematology Symp., Gent: 62.
- & OOSTENBRINK, M. (1962). Nematodes in relation to plant growth. 4. *Pratylenchus penetrans* (Cobb) on orchard trees. *Neth. J. agric. Sci.* 10: 286-297.
- (1968). Replant diseases of apple in the Netherlands. *Meded. LandbHogeschool Wageningen* 68 (13): 105 p.
- HOLLIS, J. P., WHITLOCK, L. S., ATKINS, J. G. & FIELDING, M. J. (1959). Relations between nematodes, fumigation and fertilization in rice culture. *Pl. Dis. Repr* 43: 33-40.
- (1962). A survey of plant parasitic nematodes and their control in Kenya. *Pl. Prot. Bull. F.A.O.* 10: 97-106.
- & RODRIGUEZ, K. R. (1966). Rapid kill of nematodes in flooded soil. *Phytopathology* 56: 1015-1019.
- HORN, N. L., MARTIN, W. J., WILSON, W. F. & GIAMALVA, M. J. (1956). The relation of nematodes to strawberry culture in Louisiana. *Pl. Dis. Repr* 40: 790-797.
- HURST, R. H. & FRANKLIN, M. T. (1938). Field experiments in Bedfordshire on the chemical

- treatment of soil infected with the potato eelworm *Heterodera schachtii*, during 1936-1937. J. Helminth. 16: 33-46.
- HUTCHINSON, M. T. (1959). Crop rustling nematodes cost \$ 15 million yearly. New Jers. Agric. 41: 9-12.
- , RED, J. P., STREU, H. T., EDWARDS, A. A. & SCHROEDER, P. H. (1961) Plant parasitic nematodes of New Jersey. Bull. New Jers. agric. Exp. Stn 796: 33 p.
- JACKS, H. & SMITH, H. C. (1952). Soil disinfection. 12. Effect of fumigants on growth of soil fungi in culture. N. Z. J. Sci. Technol. 33, Sect. A: 69-73.
- JENSEN, H. J., HOPPER, W. E. R. & PAGE, G. E. (1964). Control of stubby root nematode in Oregon onion plantings by soil fumigation. Pl. Dis. Reprtr 48: 225-227.
- JOHNSON, M. O. & GODFREY, G. H. (1932). Chloropicrin for nematode control. Ind. Engng. Chem. News Edn 24: 311-313.
- JOHNSON, J. (1946). Soil steaming for disease control. Soil Sci. 61: 83-91.
- JOHNSTON, T. M. (1957). Further studies on microbiological reduction of nematode population in water-saturated soils. Phytopathology 47: 525-526.
- KÄMPFE, L. (1962). Zur Wirkung von Ultraschall auf zystenbildende und freilebende Nematoden. Nematologica 7: 164-172.
- KANTZES, J. G. & MORGAN, O. D. (1962). Comparison of nematodes for control of root-knot nematodes on strawberries. Phytopathology 52: 164.
- KINCAID, R. R. & VOLK, G. M. (1949). Soil fumigation for cigar-wrapper tobacco in Florida. Phytopathology 39: 11.
- KITTLESON, A. R. (1952). A new class of organic fungicides. Science, N.Y. 115: 84-86.
- KOFFLER, H., MALLET, G. E. & ADYE, J. (1957). Molecular basis of biological stability to high temperatures. Proc. natn. Acad. Sci. U.S.A. 43: 464-477.
- KOLENBRANDER, G. J. (1969). Voorlopig N-bemestingsadvies na een DD-behandeling. De Buffer 15: 54-60, 108.
- KOOISTRA, G. (1964). The effect of DD on grass and clover in sterilized soil. Neth. J. Pl. Path. 70: 81-84.
- KRUSBERG, L. R. (1965). Fatty acids in plant-parasitic nematodes. Phytopathology 55: 1065.
- (1968). Significance of enzyme systems in relation to pathogens and control. 1. Int. Congr. Pl. Path. (Lond. 1968).
- KÜHN, J. (1881). Die Ergebnisse der Versuche zur Ermittlung der Ursache der Rübenmüdigkeit und zur Erforschung der Natur der Nematoden. Ber. physiol. Lab. landw. Inst. Univ. Halle. 3: 153 p.
- KUIPER, K. & LEEUW, W. P. DE (1963). Landbouwpoeiderkalk als nematocide, Meded. Landb-Hoogeschool. OpzoekStns Gent 28: 618-622.
- LANGE, W. H. (1947). Sugar-beet insects and nematodes and their control. Spreckels Sug. Beet Bull. 11: 1-8.
- LEAP, N. (1955). King of pests. The Saturday Evening Post, Oct. 15: (53), 142, 144, 146-147.
- LAWRENCE, W. J. C. (1956). Soil sterilization. George Allen and Unwin, Lond.: 171 p.
- LEAR, B. & JACOB, F. C. (1955). Electrical tests on nematodes. Calif. Agric. 9(10): 9.
- (1956). Results of laboratory experiments with Vapam for control of nematodes. Pl. Dis. Reprtr 40: 847-852.
- , JOHNSON, D. E., MIYAGAWA, S. T. & SCIARONI, R. H. (1965). Response of Brussels sprouts to applications of nematocides in transplant water for control of sugarbeet nematode and cabbage root nematode. Pl. Dis. Reprtr 49: 900-902.
- LECLERG, E. L. (1964). Crop losses due to plant diseases in the United States. Phytopathology 54: 1309-1313.
- LEMBRIGHT, H. W. (1959). New uses of chloropicrin as a soil fungicide. Phytopathology 49: 113.
- LEVITT, J. (1956). The hardness of plants. Acad. Press Inc., N.Y.: 278 p.
- LOWNSBERY, B. F. & SHER, S. A. (1957). Soil fumigation for control of the root lesion disease of walnuts. Phytopathology 47: 527-528.
- MACHMER, J. H. (1949). Soil fumigation for the control of the root-knot nematode in peach, fig and grape plantings. Phytopathology 39: 498.

- MAESENEER, J. DE (1967). Nematicide werking van kopersulfaat. 19. Int. Symp. Phytat.-Phytopharm., Gent: 559-564.
- MARKS, C. F., CASTRO, C. E. & THOMASON, I. J. (1966). Uptake of ethylene dibromide by nematodes. *Phytopathology* 56: 888.
- MARLATT, R. B. & ALLEN, R. M. (1959). Control of tomato root-knot in Arizona with fumigants. *Pl. Dis. Repr* 43: 309.
- MARTIN, J. P., ALDRICH, D. G., MURPHY, W. S. & BRADFORD, G. R. (1953). Effect of soil fumigation on growth and chemical composition of citrus plants. *Soil Sci.* 75: 137-151.
- MATTHEWS, D. J. (1919). Report of the work of W. B. Randall research assistant. *Rep. Exp. Res. Stn., Cheshunt* 5: 18-21.
- MCBETH, C. W. (1951). Observation on repeated applications of DD (dichloropropene-dichloropropane). *Pl. Dis. Repr* 35: 243-245.
- (1954). Some practical aspects of soil fumigation. *Pl. Dis. Repr Suppl.* 227: 95-97.
- MCCAIN, A. H., HOLTZMANN, O. V. & TRUJILLO, E. E. (1967). Concentration of *Phytophthora cinnamomi* chlamidospores by sieving. *Phytopathology* 57: 1134-1135.
- MCCLELLAN, W. D., CHRISTIE, J. R. & HORN, N. L. (1947). Effect of temperature and moisture on the efficacy of soil fumigants. *Phytopathology* 37: 440.
- , WILHELM, S. & GEORGE, A. (1955). Incidence of *Verticillium* wilt in cotton not affected by root-knot nematodes. *Pl. Dis. Repr* 39: 226-227.
- MELCHERS, L. E. (1919). Methods of steam sterilization of soil for controlling nematodes. *Phytopathology* 9: 294-296.
- MESSING, J. N. L. (1965). Extractable aluminium in steam-sterilized soils. *Nature, Lond.* 207: 439-440.
- MILES, H. W. & TURNER, W. H. (1928). On the control of the root-knot eelworm *Heterodera radiculicola* Müller. *J. Helminth.* 6: 59-76.
- MILLER, P. M. (1961). Improving growth of fruit trees by treatment with nematocides and fungicides at time of planting. *Pl. Dis. Repr* 45: 42-44.
- & WAGGONER, P. E. (1962). Control of soil-borne fungi and incidence of *Pratylenchus penetrans*. *Phytopathology* 52: 926.
- MILLER, P. R. (1957). Control of *Fusarium* wilt and root-knot nematode of tobacco. *Agric. Chem.* 12 (4): 52, 54-55.
- (1958). Control of root-knot nematodes by soil fumigation. *Agric. Chem.* 13 (10): 66-67.
- MINTON, E. B., CAIRNS, E. J. & SMITH, A. L. (1960). Effects of soil fumigants on the occurrence of nematodes in field bins. *Pl. Dis. Repr* 44: 479-483.
- MORGAN, O. D. (1957). Control of *Fusarium* wilt and root-knot nematode of tobacco with soil fumigants. *Pl. Dis. Repr* 41: 27-32.
- & JEFFERS, W. F. (1957). The effects of fumigation and heat treatment on root-knot nematode and black root rot of strawberries. *Pl. Dis. Repr* 41: 825-831.
- (1958). Observations on fumigation of tobacco soils. *Pl. Dis. Repr* 42: 316-317.
- MORITA, H. T. & GODFREY, G. H. (1933). The thermal death point of *Heterodera radiculicola* in relation to time. *Phytopathology* 23: 260-270.
- MORTON, D. J. (1959). The use of a granular nematocide applied at listing in controlling cotton root-knot. *Pl. Dis. Repr* 43: 248-252.
- MOTSINGER, R. E. (1961). Evaluation of an experimental nematicide 0,0-diethyl 0-2-Pyrazinyl phosphorothioate. *Pl. Dis. Repr* 45: 335-340.
- MULLIN, R. S. (1952). Control of cracking in sweet potato by soil fumigation. *Phytopathology* 42: 15.
- MURANT, A. F. & TAYLOR, C. E. (1965). Treatment of soil with chemicals to prevent transmission of tomato blackring and raspberry ringspot virus by *Longidorus elongatus* (de Man). *Ann. appl. Biol.* 55: 227-237.
- MYERS, R. F. & DROPKIN, V. H. (1959). Impracticability of control of plant-parasitic nematodes with ionizing radiations. *Pl. Dis. Repr* 43: 311-313.
- (1960). The sensitivity of some plant-parasitic and free-living nematodes to gamma and X-irradiation. *Nematologica* 5: 56-63.
- NELLER, J. R. & ALLISON, R. V. (1935). A machine for the subsurface treatment of soils with

- chloropicrin and with carbonbisulfide for nematode control under field conditions. *Soil Sci.* 40: 173-179.
- NEWHALL, A. G. & LEAR, B. (1948). Field trials in New York with ethylenedibromide and DD mixture against the root-knot nematode. *Phytopathology* 38: 19-20.
- NIELSEN, L. W. & SASSER, J. N. (1959). Control of root-knot nematodes affecting Porto Rico sweet potatoes. *Phytopathology* 49: 135-140.
- OAKES, J. Y., BOLICH, C. N., MELVILLE, R. D., FIELDING, M. J. & HOLLIS, J. P. (1956). A preliminary report on soil fumigation for control of parasitic nematodes on corn at Curtis, Louisiana. *Pl. Dis. Reprtr* 40: 853-854.
- O'BANNON, J. H. & REYNOLDS, H. W. (1962). Citrus tree decline caused by *Tylenchulus semipenetrans* and tree response following chemical treatment. *Phytopathology* 52: 746.
- & —, (1963). Response of navel orange trees to a post planting application of dibromochloropropane for control of the citrus nematode. *Pl. Dis. Reprtr* 47: 401-404.
- O'BRIEN D. G., GEMMELL A. R., PRENTICE, I. W. & WYLIE, S. M. (1939). Field experiments in Ayrshire on control of *Heterodera schachtii* by the use of chloroacetates. *J. Helminth.* 17: 41-50.
- OOSTENBRINK, M. (1950). Het aardappelaaltje (*H. rostochiensis* Wollenweber), een gevaarlijke parasiet voor de eenzijdige aardappelcultuur. *Versl. Meded. plziektenk. Dienst Wageningen* 115: 230 p.
- (1954). Over de betekenis van vrijlevende wortelaaltjes in land- en tuinbouw. *Versl. Meded. plziektenk. Dienst Wageningen* 124: 196-233.
- (1957). Nematoden in verband met de vruchtbaarheid van de grond. *Overdr. Plziektenk. Dienst Wageningen* 130: 21 p.
- (1958). Grondontsmetting en pH. *Meded. LandbHoogesch. OpzoekStns Gent* 23: 628-635.
- (1959). Wachstumssteigerung durch Bodenbehandlung mit Nematiziden. *Mitt. biol. BundAnst. Land- u. Forstw.* 97: 117-120.
- (1960). Estimating nematode populations by some selected methods. In: *Nematology*, J. N. Sasser & W. R. Jenkins, Univ. N. Carolina Press, Chapel Hill: 85-102.
- & HOESTRA, H. (1961). Nematode damage and 'specific sickness' in Rosa, Malus and Laburnum. *Tijdschr. PlZiekt.* 67: 264-272.
- (1961). Nematodes in relation to plant growth. 3. *Pratylenchus penetrans* (Cobb) in tree crops, potatoes and red clover. *Neth. J. agric. Sci.* 9: 188-209.
- (1964). Harmonious control of nematode infestation. *Nematologica* 10: 49-56.
- (1966). Major characteristics of the relations between nematodes and plants. *Meded. LandbHogesch. Wageningen* 66 (4): 46 p.
- OTEIFA, B. A. (1952). Potassium nutrition of the host in relation to infection by a root-knot nematode *Meloidogyne incognita*. *Proc. helminth. Soc. Wash.* 19: 99-104.
- (1965). Elements of economic plant nematology. *Scient. Notes Fac. Agric. Cairo Univ.*: 99 p.
- OWENS, R. G. & ELLIS, D. E. (1951). The efficacy of certain chemical soil treatments against meadow nematodes. *Phytopathology* 41: 123-126.
- PARKINSON, D. & WILLIAMS, S. T. (1961). A method for isolating fungi from soil microhabitats. *Pl. Soil* 13: 347-355.
- PARRIS, G. K. (1945). The nematocidal and fungicidal value of DD-mixture and other volatile soil fumigants. *Phytopathology* 35: 771-780.
- (1946). Use of DD mixture permits two crops of watermelons per year in breeding program. *Phytopathology* 36: 408.
- (1958). Soil fumigants and their use. *Pl. Dis. Reprtr* 42: 273-278.
- PEACHEY, J. E. & WINSLOW, R. D. (1962). Effects of soil treatments on populations of soil nematodes and on carrot crops grown for two years after treatment. *Nematologica* 8: 75-79.
- & CHAPMAN, M. R. (1966). Chemical control of plant nematodes. *Tech. Commun. Commonw. Bur. Helminth.* 36: 199 p.
- PETERS, B. G. (1948). Potato root eelworm, DD and soil sterilization. 2. Results for 1946. *J. Helminth.* 22: 128-138.

- PETERS, B. G. (1949). Potato root eelworm, DD and soil sterilization. *J. Helminth.* 23: 73–88.
- (1953). Trial of ammoniacal gas liquor against potato root eelworm. *Pl. Path.* 2: 65–68.
- PIEROH, E. A., WERRES, H. & RASCHKE, K. (1959). Trapex—ein neues Nematizid zur Bodenentseuchung. *Anz. Schädlingsk.* 32: 183–189.
- PITCHER, R. S. (1965). Interrelationships of nematodes and other pathogens of plants. *Helminth. Abstr.* 34: 1–17.
- POTTER, H. S. & MORGAN, O. D. (1956). Nemagon control of root-knot nematode on strawberries. *Pl. Dis. Repr* 40: 187–189.
- (1919). Nitrification in Egyptian soils. *J. agric. Sci., Camb.* 9: 216–236.
- PRESCOTT, J. A. (1919). Nitrification in Egyptian soils *J. agric. Sci. Camb.* 9: 216–236.
- (1920). A note on the sheraqi soils of Egypt. A study in partial sterilization. *J. agric. Sci., Camb.* 10: 177–181.
- RADEWALD, J. D., PAULUS, A. O. & HOFFMAN, E. C. (1964). Effect of preplant soil fumigation on stubby-root nematode control on alfalfa in southern California. *Phytopathology* 54: 1436.
- RANKIN, H. W. & GOOD, J. M. (1959). Effect of soil fumigation on the prevalence of southern blight on tomatoes. *Pl. Dis. Repr* 43: 444–445.
- (1961). Control of pathogenic fungi, nematodes, and weeds by preplanting chemical treatments. *Phytopathology* 51: 644.
- RASKI, D. J. & ALLEN, M. W. (1948). Sugar-beet nematode; identification and recommendations for control of the pest in California fields. *Calif. Agric.* 2 (11): 8, 16.
- & —, (1953). Control of root-knot nematode on cotton. *Pl. Dis. Repr* 37: 193–196.
- (1953). Methods of detecting and investigating plant-parasitic nematodes. *Phytopathology* 43: 259–263.
- (1954). Soil fumigation for the control of nematodes on grape replants. *Pl. Dis. Repr* 38: 811–817.
- (1956). *Pratylenchus penetrans* tested on strawberries grown in black root rot soil. *Pl. Dis. Repr* 40: 690–693.
- (1959). Concept and problems of nematology. In: *Plant Pathology, Problems and Progress 1908–1958*, C. S. Holton, G. W. Fisher, R. W. Fulton, H. Hart and S. E. A. McCallen Univ. Wis. Press: 384–394.
- & LEAR, B. (1962). Influence of rotation and fumigation on root-knot nematode populations on grape replants. *Nematologica* 8: 143–151.
- REYNOLDS, H. W. & HANSON, R. G. (1957). Rhizoctonia disease of cotton in presence or absence of the cotton root-knot nematode in Arizona. *Phytopathology* 47: 256–261.
- RHOADES, H. L. (1961). A preliminary report on two experimental soil fumigants. *Pl. Dis. Repr* 45: 54–57.
- (1964). Effect of hot water treatment of seed tubers and soil fumigation for control of root-knot on yield of caladiums. *Pl. Dis. Repr* 48: 568–571.
- RICHARDSON, H. H. & JOHNSON, A. C. (1935). Studies of methyl bromide in greenhouse and vault fumigation. *Tech. Bull. U. S. Dep. Agric.* 853: 1–20.
- ROHDE, R. A. (1960). Acetylcholinesterase in plant-parasitic nematodes and an anticholinesterase from asparagus. *Proc. helminth. Soc. Wash.* 27: 121–124.
- RUEHLE, J. L. & SASSER, J. N. (1964). Additional data on results of nematocidal fumigation in pine plots in North Carolina. *Pl. Dis. Repr* 48: 534–536.
- RUSSELL, E. J. & BUDDIN, W. (1914). The action of antiseptics in increasing the growth of crops in soil. *J. Soc. chem. Ind., Lond.* 32: 1136–1142.
- SAYRE, R. M. (1963). Effects of rapid cooling to subzero temperatures on *Meloidogyne hapla* and *M. incognita*. *Phytopathology* 53: 888.
- SCHMITT, C. G. (1948). Chemicals for control of the golden nematode of potatoes. *Phytopathology* 38: 23.
- SEINHORST, J. W. (1950). De betekenis van de toestand van de grond voor het optreden van aantasting door het stengelaaltje (*Ditylenchus dipsaci* (Kühn) Filipjev). *Tijdschr. PZiekt.* 56: 289–348.
- (1968). Mortality of plant parasitic nematodes shortly after removal of the host plant.



- Rep. 8. int. Symp. Pl. Nematology (Antibes 1965): 41-42.
- SHARMA, R. D. (1971). Studies on the plant parasitic nematode *Tylenchorhynchus dubius*. Meded. LandbHogeschool Wageningen 71-1: 154 p.
- SHEPHERD, C. J. (1952). Taining of tobacco by a dichloropropene-dichloropropane soil fumigant. Nature, Lond. 170: 1073-1074.
- SHER, S. A. (1957a). A disease of roses caused by a root-lesion nematode, *Pratylenchus vulnus*. Phytopathology 47: 703-706.
- (1957b). Response of roses to field fumigation for lesion nematode, *Pratylenchus vulnus*. Phytopathology 47: 31.
- , THOMASON, I. J. & McCASLIN, R. L. (1958). Chisel application of methylbromide for root-knot nematode control. Pl. Dis. Repr 42: 288-290.
- SLOOTWEG, A. F. G. (1954). Wortelrot in lelies en in narcissen. Theorie en Practijk, Lisse (1954) 36: 5-8.
- SMART, G. C., LOCASCIO, S. J. & RHOADES, H. L. (1967). Root-knot nematode control on strawberry. Nematologica 13: 152-153.
- SMEDLEY, E. M. (1936). The action of certain halogen compounds on the potato eelworm, *Heterodera schachtii*. J. Helminth. 14: 11-20.
- (1939). Experiments on the use of isothiocyanates in the control of the potato strain of *Heterodera schachtii* (Schmidt). J. Helminth. 17: 31-38.
- (1938). Experiments to determine the relative toxicity of ammonium chloroacetate and related chemicals to the potato eelworm (*Heterodera schachtii*). J. Helminth. 16: 177-180.
- SMITH, T. E. (1947). DD mixture as a soil treatment for bacterial wilt on tobacco. Phytopathology 37: 371.
- SNEDECOR, G. W. (1956). Statistical methods (5. Edn). Iowa St. Univ. Press, Ames, Iowa: 534 p.
- SONNEVELD, C. (1968). The manganese condition of the soil and the fixation of manganese in lettuce on steamed soil. Meded. Dir. Tuinb. 31: 507.
- SOUTHEY, J. F. & SAMUEL, G. G. (1954). Potato root eelworm. 1. A review of the present position. 2. Research in progress. Pamph. nat. agric. advis. Serv.: 1-13.
- SPENCER, E. Y. (1968). Guide to the chemicals used in crop protection (5. Edn). Can. Dept. Agric.: 483 p.
- SPURR, H. W. (1966). Nematode cholinesterases and their inhibition by nematocidal carbamates. Phytopathology 56: 902-903.
- STANILAND, L. N. (1953). Hot water treatment of strawberry runners. Pl. Path. 2: 44-49.
- STARK, F. L., SMITH, J. B. & HOWARD, F. L. (1939). Effect of CP fumigation on nitrification and ammonification in soil. Soil Sci. 48: 433-442.
- , LEAR, B. & NEWHALL, A. G. (1944). Comparison of soil fumigants for the control of the root-knot nematode. Phytopathology 34: 954-965.
- & LEAR, B. (1947). Miscellaneous greenhouse tests with various soil fumigants for the control of fungi and nematodes. Phytopathology 37: 698-711.
- STEELE, A. E. & GOOD, J. M. (1958). Evaluation of several nematocides for control of sting nematodes on Lima beans. Pl. Dis. Repr 42: 1284-1287.
- STEINER, G. (1953). Changes in basic concepts in plant nematology. Pl. Dis. Repr 37: 203-205.
- STELMACH, Z. (1959). Bromine retention in some soils and uptake of bromine by plants after soil fumigation. Soil Sci. 88: 61-66.
- STEMERDING, S. (1963). Een mixer-wattenfiltermethode om vrijbeweeglijke endoparasitaire nematoden uit wortels te verzamelen. Versl. Meded. plziektenk. Dienst Wageningen 141: 170-175.
- STOJANOV, D. (1962). Nekotorye itogi izučenija fitonematod v Bolgarii. Trudy 5. vses. Soveshch. Fitogel'mintologov Samarkand: 269-284.
- SUTT, R. F., DUCHARME, E. P. & FELDMAN, A. W. (1961). Effectiveness of DBCP and fungicides for the control of *Radopholus similis* on citrus trees. Pl. Dis. Repr 45: 62-66.
- SVEŠNIKOVA, N. M. (1965). Nematodičny. In: "Osnovnye itogi ispytaniy pestičidov v 1963 godu". Bjull. 5: 131-143.
- TAM, R. K. & CLARK, H. E. (1943). Effect of chloropicrin and other soil disinfectants on the nitrogen nutrition of the pineapple plant. Soil Sci. 56: 245-261.

- TAM, R. K. (1945). The comparative effects of 50-50 mixture of 1-3-dichloropropene, 1-2-dichloropropane (DD mixture) and of chloropicrin on nitrification in soil and on the growth of pineapple plants. *Soil Sci.* 59: 191-205.
- TARJAN, A. C. (1948). The inefficacy of ethylene chlorobromide as a therapeutic agent in the treatment of gardenias infected with the root-knot nematode. *Phytopathology* 38: 845-847.
- (1950a). Investigations of meadow nematodes attacking boxwood, and the therapeutic value of sodium selenate as a control. *Phytopathology* 40: 1111-1124.
- (1950b). A consideration of mineral nutrition of boxwood in relation to infection by meadow nematodes, *Pratylenchus* spp. *J. Wash. Acad. Sci.* 40: 157-160.
- TAYLOR, A. L. & MCBETH, C. W. (1940). Preliminary tests of methylbromide as a nematocide. *Proc. helminth. Soc. Wash.* 7: 94-96.
- & —, (1941a). A practical method of using methylbromide as a nematocide in the field. *Proc. helminth. Soc. Wash.* 8: 26-28.
- & —, (1941b). Spot treatments with chloropicrin and ethylenedichloride for control of root knot. *Proc. helminth. Soc. Wash.* 8: 53-55.
- (1943). Soil fumigation with chloropicrin for control of the root-knot nematode *Heterodera marioni*. *Phytopathology* 33: 1166-1175.
- & MCBETH, C. W. (1947). The effect of soil fumigation on growth and yield of peach trees. *Phytopathology* 37: 437.
- (1961). The effect of nematodes on crop yields in the United States. 6th Int. Nematology Symp., Gent: 80.
- (1967). Introduction to research on plant nematology. FAO, Rome: 133 p.
- TAYLOR, G. S. (1956). Tobacco seed bed treatments for control of weeds and soil borne pathogens. *Phytopathology* 46: 242.
- THAMES, W. H. & LANGLEY, B. C. (1964). Chemical control of *Meloidogyne arenaria* on Spanish peanuts in Texas. *Phytopathology* 54: 626.
- THOMAS, P. R., WIGGELL, P. & MOYSE, W. J. (1958). Observations on eelworm control in Narcissus in the Isles of Scilly. *Pl. Path.* 7: 49-50.
- THOMASON, I. J. (1959). Chisel application of methyl bromide for control of root-knot nematode and Fusarium wilt. *Pl. Dis. Reprtr* 43: 580-583.
- , RADEWALD, J. D., WELCH, N. C. & MCKINNEY, H. E. (1964). Onion yield increases following application of nematocides to control stubby-root, root-knot and lesion nematodes. *Pl. Dis. Reprtr* 48: 409-413.
- THORNE, G. & JENSEN, V. (1946). A preliminary report on the control of sugar-beet nematode with two chemicals, DD and Dowfume W 15. *Proc. Am. Soc. Sug. Beet Technol.* 4: 322-326.
- & SCHIEBER, E. (1962). American dagger nematode (*Xiphinema americanum*) on coffee in Guatemala, with suggestions for nematode control in nurseries. *Pl. Dis. Reprtr* 46: 857.
- TILLET, E. R. (1964). The effect of different fumigants on the mineralization of soil nitrogen. *Rhod. J. agric. Res.* 2: 13-16.
- TRIFFITT, M. J. (1929). Preliminary researches on mustard as a factor inhibiting cyst-formation in *Heterodera schachtii*. *J. Helminth.* 7: 81-92.
- & HURST, R. H. (1935). On the thermal death-point of *Heterodera schachtii*. *J. Helminth.* 13: 219-223.
- VALLEGA, J. & CHIARAPPA, L. (1964). Plant disease losses as they occur worldwide. *Phytopathology* 54: 1305-1308.
- VECHT, J. VAN DER (1950). De plagen van de cultuurgewassen in Indonesie. N.V. Uitgeverij W. van Hoeve, 's Gravenhage/Bandoeng: 16-42.
- VOLODOCOVIČ, S. D. & ČECALINA, V. I. (1967). Chemičeskie sredstva bor'by s nematodami. In: *Nematodnye Bolezni Rastenii*, Sel'khozgiz, N.M. Sveshnikova, Kolos, Mosk.: 200-208.
- WALKER, J. T. (1960). The effect of hot water at different temperature on larvae of various species of *Meloidogyne*. *Phytopathology* 50: 658.
- (1962). The sensitivity of larvae and eggs of *Meloidogyne* species to hot water treatments.

Nematologica 7: 19-24.

- WATSON, J. R. (1924). Control of root-knot nematodes on Florida truck farms. J. econ. Ent. 17: 225-230.
- WELLE, H. B. A. & BIJLOO, J. D. (1965). The systemic nematocidal activity of physostigmine and structural analogues. Meded. LandbHoogesch. OpzoekStns Gent 30: 1417-1428.
- WENSLEY, R. N. (1956). The peach replant problem in Ontario. 4. Fungi associated with replant failure and their importance in fumigated and nonfumigated soils. Can. J. Bot. 34: 967-981.
- WILHELM, S. (1965). *Pythium ultimum* and the soil fumigation growth response. Phytopathology 55: 1016-1020.
- , GEORGE, A. & PENDERY, W. (1967). Zinc deficiency in cotton induced by chloropicrin-methylbromide soil fumigation to control *Verticillium* wilt. Phytopathology 57: 103.
- WILSON, J. D. (1956). Control of root-knot on carrot, celery, and onion in muck soil by EDB and DD. Phytopathology 46: 31.
- & HEDDEN, O. (1964). Effect of repeated application of nematocides on vegetable yields and nematode populations in muck soil. Pl. Dis. Repr 48: 698-702.
- WINFREE, J. P. & COX, R. S. (1958). Comparative effects of fumigation with chloropicrin and methylbromide on mineralization of nitrogen in Everglades peat. Pl. Dis. Repr 42: 807-810.
- WOESTUNE N. VAN DE, & BRANDE J. VAN DEN (1960). Gevoeligheid van plantenparasiterende aaltjes voor radioactieve straling. Meded. LandbHoogesch. OpzoekStns Gent 25: 1057-1064.
- WOOD, F. C. & GOODEY, J. B. (1957). Effects of gamma-ray irradiation on nematodes infesting cultivated mushroom beds. Nature, Lond. 180: 760-761.
- YOUNG, P. A. (1964). Control of corn nematodes with Vorlex and DD. Pl. Dis. Repr 48: 122-123.
- YOUNG, V. H. & SMITH, H. E. (1953). Soil fumigation for the control of root-knot and *Fusarium* wilt of cotton. Arkans. Fm Res. 2: 2-3.
- ZENTMYER, G. A. & KENDRICK, J. B. (1949). Fungicidal action of volatile soil fumigants. Phytopathology 39: 864.