AN ABSTRACT OF THE THESIS OF

Jeanette Anne McKewan for the degree of Master of Science
in Botany and Plant Pathology presented on August 10, 1979

Title: THE BIOLOGY AND PATHOGENICITY OF A SHEATH NEMATODE Hemicycliophora similis THORNE, 1955

Abstract approved: Redacted for Privacy

Harold J. Jensen

A study, using greenhouse and laboratory experiments, was made of Hemicycliophora similis from Bandon, Oregon to determine: identity and morphometrics, time and development of a life cycle, host plant preferences and optimum environmental conditions for reproduction and pathogenicity.

Morphometric investigations revealed discrepancies between the measurements of this nematode species and those recorded in nematological literature for H. similis. Personal communications with R. P. Esser, H. J. Jensen and B. M. Zuckerman suggested the variations noted do not necessitate describing a new species. For purposes of this research the nematode is named Hemicycliophora similis.

Life history investigations were conducted to determine the number of days required to complete a life cycle and the various stages as the nematode developed from egg to egg. Eggs mature to
first-stage larvae, without sheath or spear, in 3-5 days and hatch as second-stage larvae in 8-10 days. Second-stage larvae inoculated onto carrot seedlings matured into third-stage larvae in 12-15 days. Fourth-stage larvae appeared in 25-28 days, and adults were present in 35-38 days. Embryonating eggs were observed lodged within an incompletely shed fourth-stage cuticle attached to adult females and within the normal sheath surrounding the adult. One molting specimen was found with two eggs lodged within the fourth-stage cuticle. Such eggs were seen to develop to second-stage larvae; however, none were observed over a long enough time period to determine if larvae hatched and emerged free of the cuticular confinement.

Evaluated as hosts were beet, broccoli, carrot, corn, dill, pea, radish, rutabaga, tomato and turnip. Carrot, tomato and turnip supported the highest increase in population; no substantial increase was observed on beet, broccoli, corn and dill. The nematode formed galls consisting of large multibranched growths and reproduced on many of the plant species tested. Galling of seedling roots was most severe on carrot, but slight galling was observed on beet, rutabaga and turnip. Multibranching was observed with varying severity in all hosts. Germination of carrot seeds was reduced substantially when grown in sandy soil infested with H. similis. Slight germination reductions occurred with all hosts tested except corn and pea. Adults and larvae were observed feeding behind root tips and on nematode induced galls.
Root discoloration was noted on several plants.

To evaluate temperature and soil optima, rooted cranberry cuttings, germinated cranberry seedlings and carrot seedlings were utilized. Carrot seeds were planted in sandy soils of two different textures and pH, infested with H. similis and grown in three temperature regimes: 30°C and 24°C night, 22°C day and 14°C night and 30°C day 6°C night. The optimum temperature for reproduction on carrot was 22°C day 14°C night in both soil types. A pH gradient developed in cups with the least acidic pH occurring at the base corresponding to maximum nutrient uptake by plant roots. The soil mix in which bog sand from Bandon was incorporated developed a more acidic condition presumably due to the additional organic N content which supplied ammonical nitrogen. The soil mix with Newport sand had no additional organic N and a less acidic condition developed. Maximum nematode population and root density also occurred at the base of the cup. Plants grown in infested soil had sparse multi-branched roots; no galls were observed in this experiment.

Cranberry cuttings were grown in sterile bog soil from Bandon, Oregon and inoculated with 225 nematodes/450 cc soil. The optimum temperature for nematode reproduction and plant growth was 30°C and 24°C night. Runner growth and root dry weights were not significantly effected by nematode feeding. Germinated cranberry seedlings
were subjected to similar conditions; temperature response was similar to that of cuttings. Root dry weights and runner growth of cranberry seedlings was greater in nematode inoculated treatments than non-inoculated controls: nematode feeding on cranberry roots resulted in an increase of multibranching.

This research indicated nematode feeding was detrimental to growth of a variety of plant species. High soil populations of H. similis caused severe reductions to root growth and seed germination. However, since this study was conducted in controlled conditions, it was not substantiated that this nematode caused a reduction of growth to field-grown cranberry plants. Further investigations are needed to correlate these findings with field data to determine pathogenicity on cranberry.
The Biology and Pathogenicity of a Sheath Nematode
_Hemicycliophora similis_ Thorne, 1955

by

Jeanette Anne McKewan

A THESIS

submitted to

Oregon State University

in partial fulfillment of
the requirements for the
degree of

Master of Science

Completed August 1979

Commencement June 1980
APPROVED:

Redacted for Privacy
Professor of Nematology in charge of major

Redacted for Privacy
Chairman of Department of Botany and Plant Pathology

Redacted for Privacy
Dean of Graduate School

Date thesis is presented August 10, 1979

Typed by Opal Grossnicklaus for Jeanette Anne McKewan
TABLE OF CONTENTS

INTRODUCTION 1

LITERATURE REVIEW 3

MATERIALS AND METHODS 9

- Culture of Nematodes and Plants 9
  - Soil Mixtures 9
  - Capillary Watering System 10
  - Nematode Surface Sterilization 10
  - Seed Sterilization 10
- Procedures for Data Collection 11
  - pH Measurements 11
  - Nematode Extraction 11

SECTION I: HOST RANGE STUDIES OF Hemicycliophora similis 14

- Preface 14
- Materials and Methods 14
- Results and Discussion 17

SECTION II: INFLUENCE OF ENVIRONMENTAL FACTORS ON Hemicycliophora similis AND HOST 25

- Preface 25
- Materials and Methods 25
- Nematode-Carrot Interactions 25
- Nematode-Cranberry Interactions 26
- Results 29
  - Effects on Reproduction and Pathogenicity of Nematode on Carrot (Daucus carota L.) 30
  - Effects on Reproduction and Pathogenicity of Nematode on Cranberry (Vaccinium macrocarpon Ait.) 33
- Discussion 39

SECTION III: LIFE HISTORY AND FEEDING OBSERVATIONS OF Hemicycliophora similis 46

- Preface 46
- Materials and Methods 46
Results

Egg 47
First-stage Larvae 48
Second-stage Larvae 48
Third-stage Larvae 51
Fourth-stage Larvae 52
Adult Females 52
Discussion 53

SECTION IV: TAXONOMY (MORPHOMETRICS) OF
Hemicycliophora similis 54

Preface 59
Materials and Methods 59
Results and Discussion 60

CONCLUSIONS 68

LITERATURE CITED 71

APPENDICES 76
**LIST OF FIGURES**

<table>
<thead>
<tr>
<th>Figure</th>
<th>Description</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Initial and final populations and determination of host suitability of <em>Hemicycliophora similis</em> on several test plants.</td>
<td>16</td>
</tr>
<tr>
<td>2.</td>
<td>Examples of test plant roots from sterile and nematode infested soil: A) Sterile dill, B) Infected dill, C) Sterile broccoli, D) Infected broccoli, E) Sterile beet, F) Infected beet, G) Sterile turnip, H) Infected turnip, I) Sterile rutabaga, J) Infected rutabaga, K) Sterile carrot, L) Infected carrot, M) Sterile tomato, N) Infected tomato, O) Sterile white radish, P) Infected white radish, Q) Sterile pea, R) Infected pea.</td>
<td>21</td>
</tr>
<tr>
<td>3.</td>
<td>Planting methods for cranberry: A) One replication showing capillary watering system, B) Cranberry seedlings in petri dish containers.</td>
<td>28</td>
</tr>
<tr>
<td>4.</td>
<td>Influence of temperature and soil type on population increase of <em>Hemicycliophora similis</em> on carrot.</td>
<td>31</td>
</tr>
<tr>
<td>5.</td>
<td>Influence of temperature on reproduction of <em>Hemicycliophora similis</em> and feeding effects on runner growth and root dry weight of rooted cranberry cuttings (<em>Vaccinium macrocarpon</em> Aitt).</td>
<td>35</td>
</tr>
<tr>
<td>6.</td>
<td>Influence of temperature on reproduction of <em>Hemicycliophora similis</em> and feeding effects on runner growth and root dry weight of cranberry seedlings (<em>Vaccinium macrocarpon</em> Ait.).</td>
<td>37</td>
</tr>
<tr>
<td>7.</td>
<td>Roots of cranberry seedlings grown in sterile and inoculated silica sand and bog soil, A) Roots grown in sterile silica sand, B) Multibranched roots grown in inoculated silica sand, C) Stubby roots grown in inoculated silica sand, D) Roots grown in sterile bog soil, E) Twisted roots grown in inoculated bog soil, F) Swollen root tips grown in inoculated bog soil.</td>
<td>40</td>
</tr>
</tbody>
</table>
Figure


9. Unusual occurrence of embryonating eggs lodged within the fourth larval and adult cuticle, A) Two embryonating eggs within fourth larval cuticle of a molting fourth-stage larvae, B) Two-celled egg within the fourth larval cuticle, C) Egg within shed cuticle of fourth stage-larvae, D) Embryonating egg within adult cuticle.

10. Scale drawings of heads and tails of all developmental stages of *Hemicycliophora similis*, A) Head of second-stage larvae, B) Head of third-stage larvae, C) Head of fourth-stage larvae, D) Head of adult female, E) Tail of second-stage larvae, F) Tail of third stage larvae, G) Tail of fourth-stage larvae, H) Tail of adult female.
LIST OF TABLES

<table>
<thead>
<tr>
<th>Table</th>
<th>Description</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Germination reduction, root dry weights and root symptoms of test plants in host range study.</td>
<td>18</td>
</tr>
<tr>
<td>2.</td>
<td>Final measurements of soil pH, moisture and nematode populations from carrot seedlings grown in three temperature regimes and two soil types.</td>
<td>32</td>
</tr>
<tr>
<td>3.</td>
<td>Germination percent and plant size of carrot seedlings grown in nematode infested and sterile soil under different environmental conditions.</td>
<td>34</td>
</tr>
<tr>
<td>4.</td>
<td>Runner growth, fresh top weight, root dry weight and nematode populations from cranberry seedlings grown in sterile and inoculated silica sand and bog soil.</td>
<td>38</td>
</tr>
<tr>
<td>5.</td>
<td>The number of days occurring between egg deposition and appearance of adults of Hemicycliophora similis as determined in greenhouse and laboratory investigations.</td>
<td>50</td>
</tr>
<tr>
<td>6.</td>
<td>Measurements of adult Hemicycliophora similis found in Bandon, Oregon compared with those derived by Thorne (1955) and Bryzeski (1974).</td>
<td>61</td>
</tr>
<tr>
<td>7.</td>
<td>Descriptive key to symbols used in measurements of Hemicycliophora similis.</td>
<td>62</td>
</tr>
<tr>
<td>8.</td>
<td>Measurements of Hemicycliophora similis native to cranberry bogs in Bandon, Oregon.</td>
<td>66</td>
</tr>
</tbody>
</table>
INTRODUCTION

The ectoparasitic sheath nematode *Hemicycliophora similis* Thorne, 1955, found in cranberry bogs in Bandon, Oregon was studied to gain information about its biology and pathogenicity. The nematode was first found in large numbers in soil samples submitted to the Plant Clinic of Oregon State University in October, 1972. Since then, the nematode has been maintained in flats of peppermint in the nematology greenhouse. In order to learn more about the nature and behavior of this nematode and its effect on plant growth; the interrelationships between the nematode, hosts and surrounding environment were explored to evaluate the potential importance of this nematode as a plant pest.

Certain species of *Hemicycliophora* have been reported to cause damage to economic crops. *H. arenaria* Raski, 1958 causes distinct galling of terminal and lateral root tips of rough lemon and tomato, Van Gundy and Rackham (1961). Kuiper (1959) found *H. typica* de Man 1921 to be associated with reduced growth of carrot and corresponding stubby root symptoms. *H. similis* Thorne, 1955 has been reported to cause small terminal root galls resulting in reduced root growth on blueberry and cranberry, Zuckerman (1961, 1964). Associations of large numbers of *H. similis* in the
rhizosphere of stunted and unhealthy plants in cranberry bogs near Bandon, Oregon suggest a potential pest relationship.

The aims of this investigation are to examine the life history of this nematode and determine pathogenic capabilities on a variety of hosts which support its reproduction. By making observations on the feeding process of *H. similis* on the host plants tested, the associated symptoms of parasitic activity can be described. Since there was no previous information concerning the life history of this species it seemed important to study this phase of *H. similis* biology. These findings combined with morphometric studies further characterize the species identity of this nematode. The information derived from this research will provide a more accurate evaluation of its impact on Oregon agriculture particularly cranberry production.
Formation of the nematode genus Hemicycliophora was suggested by de Man in 1921 to accommodate a single male specimen (H. typica) he found in a compost pile in a park at Bergen-op-Zoom, the Netherlands. Micoletzky in 1925 found and described 18 specimens for which he erected a new genus Procriconema and his descriptions were based on female specimens. Loos in 1948 found females of P. membranifera associated with males of H. typica in Ceylon soils. On the basis of this association he placed the females into the genus Hemicycliophora and regarded Procriconema to be a synonym. Males differ noticeably from females in this genus; they are generally rare, lack a stylet, have a degenerate esophagus, are smaller and lack the double cuticle of the female. The extra cuticle or sheath was originally considered the retention of the last larval cuticle resulting in an unfinished molt, but studies of Johnson et al. (1970) showed that this is a special formation of the cuticle occurring before each molt. There are no males associated with H. similis Thorne, 1955, the nematode studied in this investigation. A detailed historical account tracing the taxonomic development of the genus is given by Tarjan (1952).

Members of the genus Hemicycliophora are migratory ectoparasitic nematodes commonly known as 'sheath' nematodes. They
are rarely found attached to roots of the damaged plant, but can be found in surrounding soil; consequently, they were slow to be identified as causal agents of plant disease. Christie (1953) directed attention to the pathogenic capabilities of ectoparasitic nematodes and currently many are important agents in plant disease problems. Numerous studies have demonstrated that ectoparasitic nematodes incite a wide range of host responses. Damage caused by some ectoparasitic nematodes can be easily observed on roots. Conditions include absence of feeder roots, stubby roots, galling, necrotic lesions, or simply general unthriftiness of the plant caused by reduction of root systems.

The feeding process of ectoparasitic nematodes varies between species and host. Agar cultures and root observation boxes are usually considered an adequate means of studying the feeding behavior of plant parasitic nematodes. Although this system is adapted for microscopic study, it necessitates sterile conditions and does not represent the natural environment of the nematode or plant. By such observations, the behavior of nematodes reaching hosts as well as physical and chemical stimuli that attract them to host tissues have been reviewed by Jones (1960), Klinger (1965), C. D. Green (1971) and Doncaster (1971). Detailed descriptions of H. similis feeding on lettuce were made by Klinkenberg (1963) and feeding of H. arenaria on tomato by McElroy and Van Gundy (1968).
Species of *Hemicycliophora* have a flexible stylet capable of penetrating depths of 2-3 cells into root tissue. During the feeding process of *H. arenaria* an adhesive plug of polysaccharide material is formed and appears to be an attachment device securing the nematode to tomato roots, McElroy and Van Gundy (1968). This unique adaptation was also observed with *H. similis* on cranberry by Kisiel et al. (1971). The plug may also help the nematode to maintain feeding position over extended periods of time.

Feeding by *Hemicycliophora arenaria* causes distinct galling of terminal and lateral root tips. Van Gundy and Rackham (1961) determined that the root swellings occur in response to the inhibition of cell elongation at the feeding site with the concomitant acceleration of cell division. Protein accumulations and enlarged nuclei were observed within the expanded feed-cell. As feed-cells collapse, the pericycle produces new tissue which is fed upon by the nematode. Van Gundy and Rackham also contend that *H. arenaria* has evolved to an advanced stage of adaptability to the host owing to its well developed median bulb which provides for an extended salvation time, the presence of an adhesive plug and diligent selection of tissue on which it feeds. They point out limitations in this adaptation since the feeding process has a destructive effect on root development resulting in a reduction in growth of citrus and tomato. These alterations continue
as long as the nematode feeds. Normal root growth resumes when the stylet is removed as reported by the above authors.

Zuckerman (1961) observed two distinct forms of injury to cranberry roots by the feeding of *H. similis*. First, he observed that elongation of root cells on the side of the feeding site was arrested while normal growth continued on the opposite side which resulted in curved root tips. The second observation was root galling.

Khera and Zuckerman (1963) observed that the feeding of *H. similis* on ryegrass caused a cessation of root growth. The same response was observed on alfalfa, beet, broccoli, carrot, dill, radish and tomato. Bird and Jenkins (1964) reported *H. similis* to cause an overall stunting to runners and roots and a discoloring of root systems of cranberry plants. McElroy (1972) observed the formation of small terminal root galls and reduction of root growth caused by *H. similis* in laboratory experiments. He also found the nematode reduced runner formation and length as well as fresh top weight of cranberry plants. Zuckerman (1961) found *H. similis* to consistently reduce seedling vigor but noticed little effect on cranberry cuttings.

Investigations on the biology and ecology of *Hemicycliophora* spp. have resulted in finding the nematodes in various habitats associated with numerous hosts. Zuckerman et al. (1964) found *H. similis* to be the dominant species in the upper layers of Massachusetts cranberry bogs but unevenly distributed throughout
the cranberry bogs in densely infested pockets. He also reported seasonal variations in the vertical distribution of the nematode population. Dense aggregations of both sexes of *H. typica* have been recovered in sandy soils in England as well as brackish soils in Germany (Winslow, 1960). Kuiper (1959) found *H. typica* to be associated with poor growth in carrots in marine sandy soil of the Netherlands. In greenhouse inoculation trials the nematodes reproduced on carrot, iris, sugarbeet and wheat when planted in sandy soils. He also noted very few males associated with the species indicating the tendency toward parthenogenesis.

It is well documented in nematological literature that nematode feeding usually causes growth reduction in plants. Growth stimulation caused by a light infection of nematodes has also been reported. In a review by Seinhorst (1961) it is noted that certain rice varieties are well adapted to infections of *Radopholous oryzae* and when grown in the absence of the nematode will tiller excessively causing yield reduction. Enhanced growth of sugar beet in low inoculum levels of *Heterodera schachtii* has been recorded by Jones (1957). Similar stimulation was observed by Lownsberry and Peters (1955) in tobacco when grown in the presence of low population densities of *H. tobaccum*.

The physical factors in soils such as pore space, temperature and moisture and biotic factors effect nematode activity and the length
of time between each developmental stage within the life cycle of the nematode. Jones (1977) relates that coarse sandy soils favor ektoparasitic nematode activity because such soils contain more usable space than finely textured soils with a more dense structure which confines roots and nematodes to major cracks. Van Gundy and Rackham (1961) found that temperature of the soil environment directly influenced the infection process, reproduction, mobility and host invasion of Hemicycliophora arenaria. Adequate moisture is necessary for nematodes to move efficiently through the soil environment. Wallace (1958, 1963) showed that nematodes moved most efficiently in water films 2-5 μm thick which provided sufficient force to allow forward movement without slip. In thicker films they move inefficiently and in thinner ones they are unable to move at all. Biotic factors include all of the microorganisms which must compete for space and food to survive in the soil system. There are predacious and parasitic microorganisms which can attack the nematode and competitors which cause injury to host roots, such as fungi, bacteria and other nematodes. Competitors that damage the food supply or make it less suitable for nematodes may have an effect on the population. Generally information concerning life cycles has been obtained under controlled conditions and may differ from a natural occurrence in the soil.
GENERAL MATERIALS AND METHODS

The general techniques and materials used throughout this research for experimentation and data collection are outlined in this section. Materials and methods which are specific for single experiments are explained in individual sections.

Culture of Nematodes and Plants

Soil Mixtures

Greenhouse soil mix consisting of a 2:1 combination of sand and soil was autoclaved for two hours at 120°C and 15 lbs pressure. After sterilization, soil was stored in containers to cure and stabilize at least two months before potting.

To prepare infested soil, peppermint cuttings *Mentha piperita* L. var. 'Todd' and 'Mitcham' were planted in sterile greenhouse mix. When cuttings had rooted, they were drench inoculated with soil wash water containing *Hemicycliophora similis*.

Two different soil mixes were utilized for carrot studies in Section II. Mix #1 was a 1:1 combination of infested greenhouse soil and sterilized sand from Newport, Oregon with the initial pH 6.2±0.1. Mix #2 was a similar combination of infested greenhouse soil and sterile sand from Bandon, Oregon with the initial pH 6.7±0.1. Soil used for controls was a 1:1 mixture of sterile sand from Bandon or
Newport and the sterile greenhouse mix having an initial pH similar to infested mixes. Plants were fertilized twice during the growth period with a 23-19-17 water soluble fertilizer.

Capillary Watering System

Several experiments utilized modified styrofoam cups to provide a more uniform moisture regime within the small containers. The bottoms were removed and replaced with a square of cotton flannel large enough to be securely fastened to the container with a rubber band. Cups were then placed on trays which contained one cm of washed silica sand. Sand in trays was kept moist throughout the experiments.

Nematode Surface Sterilization

Nematodes were surface sterilized in 20 ppm chlorine solution for 20 minutes on a magnetic stir plate, a process developed by Chantanao and Jensen (1969), then poured through a clean 325 mesh screen and collected in a sterile watch glass. Inoculations were made in a clean work area; all tools were dipped into alcohol and flamed before each inoculation into 0.5% water agar.

Seed Sterilization

Aseptic cultures of carrot seedlings were utilized in life
history and feeding observation studies (Section III). Seeds were sterilized in 10% chlorine solution for 10 minutes on a magnetic stir plate. Seeds were then washed 4x with sterile distilled water, soaked in 0.1% aqueous mercuric chloride (Street, 1969) for 5 minutes and washed 4x with sterile distilled water and planted on 0.5% water agar.

**Procedures for Data Collection**

**pH Measurements**

The pH of sterile and infested soils was measured at the beginning and end of greenhouse experiments. A battery operated portable pH meter\(^1\) and a combination electrode was used for all measurements. Soil was placed in a 100 ml beaker previously rinsed with distilled water. Enough distilled water was added to make a thick slurry which was allowed to sit for several minutes before the probe was inserted. Initial and final measurements were the average of three readings.

**Nematode Extraction**

To determine nematode population per pot, soil was wet sieved through a gradient of mesh screens (30-100-250-325); sievings were collected and placed in Baermann funnels. The Baermann funnel

---

\(^1\)Chemtrix model 40E, manufactured by Chemtrix, Inc., Hillsboro, Oregon.
separation technique of Christie and Perry (1951) was used for all nematode extractions with some modifications. Plastic funnels containing a sample support barrier of copper wire screen (30-50 mesh) 3-5 cm from the rim, were also equipped with a short rubber hose attached to the spout at one end and sealed off at the other with a pinch clamp. Funnels were partially filled with water before a 1-ply facial tissue was placed on the wire screen. Sieving debris was carefully poured on the tissue. After 4-6 days nematodes that migrated through the tissue and into the rubber hose were drained into a 75 ml test tube by unclamping the rubber hose. After nematodes settled to the bottom of the test tube half of the contents was decanted and the remainder was poured into a counting chamber and examined with a dissecting microscope at 12.5 magnification. Turbid samples were poured through a 325 mesh screen before counting to remove excess debris. Funnels were drained again at 10 days and processed in the same manner.

Root Density

Roots free of all soil and debris were placed in an open petri dish and oven dried at 65°C for 6 days to obtain dry weights. Root dry weight was recorded to quantify root density in experiments.
Moisture Determinations

Moisture data were derived by removing three tablespoons of soil from each depth of soil measured (see carrot experiments in Section II). Soil was placed in an open petri dish, labeled, weighed and oven dried at 65°C for 7 days. Dishes and contents were re-weighed and calculations were made to determine percent of soil moisture as follows:

\[
\frac{\text{wet soil weight} - \text{dry soil weight}}{\text{wet soil weight}} \times 100 = \% \text{ Moisture}
\]
SECTION I: HOST RANGE STUDIES OF
Hemicycliophora similis

Preface

Host range studies are an effective means for determining nematode relations on a variety of plant species that may be considered in a cropping program. Seinhorst (1961) states that a plant is a host for a nematode species if the nematode can derive sufficient food to sustain growth and reproduction. On non-host plants a nematode cannot complete its life cycle although some development may occur. Therefore, a plant is not a host unless a nematode species can complete a life cycle upon or within it. Studies developed to determine host relationships of various plants for a given nematode can be helpful for devising crop rotations or other cultural control schemes. This investigation was developed to examine reproductive and pathogenic capabilities on a variety of host plants representing several plant families.

Materials and Methods

Plants representing the families Chenopodiaceae, Cruciferae, Gramineae, Labiatae, Leguminosae, Solanaceae and Umbelliferae (including beet, broccoli, carrot, corn, dill, pea, white and red radish, rutabaga, tomato and turnip) were tested to see if
Hemicycliophora similis would reproduce and form galls, multi-branched roots or other symptoms.

Seeds were planted in cups containing 150 cc of sterile or infested soil. Infested soil was obtained from two stock cultures containing different nematode population levels. The entire host range study was planted in two time intervals, the first group of plants included carrot, corn, pea, red and white radish, rutabaga, tomato and turnip. The second group of plants included beet, broccoli and dill. Soil used for the first group of test plants had an initial population of 345/150 cc soil, and soil for the second group contained about 800/150 cc soil (see Fig. 1 for the initial nematode population for each test plant). Initial nematode populations were determined by using the modified Baermann funnel technique and counting nematodes in 25 cc of extractant. A replication consisted of one cup of infested soil and one cup of sterile soil planted with an equal amount of seed. Each test plant was replicated four times and set in a randomized block design. All plants were grown in a controlled atmosphere room maintained at 30°C day and 24°C night with 12 hours illumination.

Initially, the experimental time interval was two months, to allow nematode populations to develop at least one generation. Circumstances necessitated an earlier assay of the experiment. Beet, broccoli and dill grew for 40 days and all other test plants grew for
Fig. 1. Initial and final populations and determination of host suitability of *Hemicyclophora similis* on several test plants.
58 days. At the end of the growth period, roots from each plant were washed free of soil and debris and observed with a dissecting microscope for galls and multibranched roots. Infested soil from roots and containers was wet sieved, collected and placed on Baermann funnels to obtain nematode populations. Because of extreme variability between replications of each test plant, roots of similar test plants were combined for dry weight and symptom observations. Controls were treated similarly.

**Results and Discussion**

Results of the host range study are summarized in Fig. 1. Test plants were classified as 'G' or 'N' for good or non-host. A good host supported a population increase of greater than 100% of inoculum. Those test plants in which the nematode population remained static or was reduced from the initial population were classified as non-hosts. Nematode populations on beet, broccoli and dill were examined after 40 days. The remaining host plants tested: carrot, corn, pea, red and white radish, rutabaga, tomato and turnip grew for 58 days. Results from nematode extraction procedures indicated varying populations on each test plant.

The total dry weight of roots of combined replications, % reduction of infested root dry weights, % germination and corresponding root symptoms are listed in Table 1. Statistical tests were
Table 1. Germination reduction, root dry weights and root symptoms of test plants in host range study.  

<table>
<thead>
<tr>
<th>Host</th>
<th>% reduction in germination</th>
<th>Root dry weight (g)</th>
<th>% reduction in infested root dry weight from controls</th>
<th>Root symptoms</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>infested</td>
<td>sterile</td>
<td>infested</td>
</tr>
<tr>
<td>Beet</td>
<td>75%</td>
<td>0.015</td>
<td>0.021</td>
<td>29%</td>
</tr>
<tr>
<td>Broccoli</td>
<td>15%</td>
<td>0.042</td>
<td>0.053</td>
<td>21%</td>
</tr>
<tr>
<td>Carrot</td>
<td>60%</td>
<td>0.047</td>
<td>0.779</td>
<td>94%</td>
</tr>
<tr>
<td>Corn</td>
<td>0%</td>
<td>0.729</td>
<td>1.088</td>
<td>33%</td>
</tr>
<tr>
<td>Dill</td>
<td>15%</td>
<td>0.041</td>
<td>0.049</td>
<td>16%</td>
</tr>
<tr>
<td>Pea</td>
<td>0%</td>
<td>0.953</td>
<td>0.968</td>
<td>2%</td>
</tr>
<tr>
<td>Red Radish</td>
<td>25%</td>
<td>0.215</td>
<td>0.200</td>
<td>0%</td>
</tr>
<tr>
<td>White Radish</td>
<td>25%</td>
<td>0.098</td>
<td>0.738</td>
<td>59%</td>
</tr>
<tr>
<td>Rutabaga</td>
<td>25%</td>
<td>0.013</td>
<td>0.126</td>
<td>90%</td>
</tr>
<tr>
<td>Tomato</td>
<td>15%</td>
<td>0.040</td>
<td>0.079</td>
<td>49%</td>
</tr>
<tr>
<td>Turnip</td>
<td>25%</td>
<td>0.026</td>
<td>0.342</td>
<td>92%</td>
</tr>
</tbody>
</table>

*R*oots were combined from four replications to give a total dry weight for test plants grown in infested and sterile soil.
not run on these data; since the variation due to replications was so
great any significance due to treatments would have been masked.
Therefore, only trends that were observed are reported and dis-
cussed.

The greatest reduction in root dry weights resulting from
nematode feeding occurred with carrot, rutabaga and turnip; all
were reduced by 90% or higher. White radish roots were reduced by
59% and roots of carrot were reduced by 49%. Corn roots were re-
duced by 33% and reductions of less than 30% occurred with beet,
broccoli, dill and pea. Infected roots of red radish showed no reduc-
tion, but an 8% increase occurred, possibly representing a slight
stimulation in root growth caused by nematode feeding.

Nematode feeding reduced germination in many of the test
plants. Germination of carrot seedlings was reduced by 60% com-
pared to seeds grown in sterile soil. A 25% reduction in germination
occurred with both radish varieties, rutabaga and turnip. Beet,
broccoli, dill and tomato seedling germination was reduced by 15%
and no reduction occurred with corn and pea.

Abnormalities observed on roots were galls consisting of large
multibranched growths (see Fig. 2 A-R for root symptoms). These
galls were most severe on carrot and were also observed on roots of
beet, rutabaga and turnip. Very slight multibranching of roots was
observed on broccoli, dill, both radish varieties and tomato. A
Fig. 2. Examples of test plant roots from sterile and nematode infested soil: A) Sterile dill, B) Infected dill, C) Sterile broccoli, D) Infected broccoli, E) Sterile beet, F) Infected beet, G) Sterile turnip, H) Infected turnip, I) Sterile rutabaga, J) Infected rutabaga, K) Sterile carrot, L) Infected carrot, M) Sterile tomato, N) Infected tomato, O) Sterile white radish, P) Infected white radish, Q) Sterile pea, R) Infected pea.
Figure 2.
sloughing layer of necrotic tissue produced a noticeable discoloration on infected roots of pea, turnip and red and white radish. No abnormal root symptoms appeared on corn.

Carrot, pea, both radish varieties, rutabaga, tomato and turnip were designated as good hosts. These plants developed abnormal root systems (see Fig. 2) and a population increase of over 100%. Turnip supported the highest nematode population of all hosts tested, however, pea supported the highest population with least damage to host roots. Tomato and carrot supported a substantial population increase with tomato having the lesser amount of damage to germination and roots.

Beet, broccoli, corn and dill were designated non-hosts since no substantial increase in nematode populations was detected. Since feeding symptoms occurred on beet, broccoli and dill an extended growth period may have resulted in a population increase. Information derived from these studies indicated that a variety of plant species can support population growth of *H. similis* and feeding of this nematode results in galls of multibranched growths and slight multibranching or roots.
SECTION II: INFLUENCE OF ENVIRONMENTAL FACTORS ON NEMATODE AND HOST

Preface

Under different environmental conditions pathogenicity usually influences the reproductive capacity of nematodes as well as growth development of the host. In host range studies (Section I), germination of carrot seedlings was severely restricted when planted in sandy soils infested with H. similis. Surviving carrots showed galled roots in response to nematode feeding. Zuckerman (1961) found H. similis to cause a consistent reduction in the growth of cranberry seedlings but no reduction in growth of cuttings.

Using carrot seedlings and cranberry cuttings and seedlings as host plants, this study evaluates effects of feeding on top and root growth of host plants.

Materials and Methods

Nematode-Carrot Interactions

Carrot seeds were planted in two different mixtures of infested greenhouse soil in 450 ml styrofoam cups modified to provide a capillary watering system (see General Materials and Methods). Twelve cups were placed on each tray, nine filled with infested soil and three with sterile soil for controls.
Temperature regimes used for this experiment were: a control room at 30°C day and 24°C night ±2°C, a Sherer\textsuperscript{2} model 25-7HL Controlled Environment Chamber set at 22°C day and 24°C night ±1°C and a Percival\textsuperscript{3} walk-in growth chamber at 20°C day 6°C night ±1°C.

After a two-month growth period each cup was divided horizontally into three equal depths and the soil was processed for nematode populations, moisture and pH. Root densities were examined visually and all plants were observed for galling and multibranching.

**Nematode-Cranberry Interactions**

Cranberry seedlings and rooted cuttings were used in similar trials to evaluate environmental effects on reproductive rate and pathogenic expression. Cranberry runners collected from the cranberry bogs at Bandon, Oregon, were cut into 15-20 cm lengths and rooted in vermiculite for 3 months. Modifying procedures outlined by Devlin and Karczmarczyk (1974), cranberry seeds, extracted from whole berries obtained in fall 1978 from the Bandon bogs, germinated in 8-10 days at 25°C with 12 hours illumination.

Germinated seedlings and cuttings were planted in sterile bog

\textsuperscript{2}Sherer-Gillett Co., Marshall, Michigan.

\textsuperscript{3}Percival Manufacturing Co., Boone, Iowa 50036.
soil, initial pH 4.9, obtained from Bandon, Oregon. To insure adequate fertility to cranberries throughout the experiment, soil was mixed with 0.34 g/l of a urea-formaldehyde urea mixture, as reported by Somogyi et al. (1963) (see Appendix A).

Sterile bog soil was placed in 450 ml styrofoam cups, modified to provide a capillary watering system, and planted with germinated cranberry seedlings or rooted cuttings. Two cups planted with seeds or cuttings, placed in a sand lined 9"x9" aluminum baking tray, made up each replication (see Fig. 3A). Plants were grown in 30°C day and 24°C night with 12 hours illumination for two months to develop root systems. After the root development period, half of the replications of cranberry cuttings and seedlings were placed in 22°C day and 14°C night; inoculations of 225 nematodes were made in one cup per tray. To reduce variability in the inoculating procedure, trays in both temperatures were inoculated simultaneously. Plants were measured at the time of inoculation and at the end of the growing period.

At the end of the growth period, soil from both seedlings and cuttings was assayed for nematode populations and plants were analyzed for runner growth and root dry weight.

In another study cranberry seedlings were planted in plastic petri dishes (three seedlings per dish) containing sterile silica sand or sterile bog soil. Holes were drilled in lids to allow for runner growth (see Fig. 3B). Inoculations of 150 nematodes per plant were made by inserting a 2 ml automatic pipet into holes. Runners were
Figure 3. Planting methods for cranberry: A) one replication showing capillary watering system, B) cranberry seedlings in petri dish containers.
measured at the time of inoculation. An equal number of uninoculated plants remained as checks. Plants were watered through holes with complete Hoagland's solution (see Appendix A).

After a two-month growing period in 25°C±1°C with 12 hours illumination, measurements of nematode populations, runner growth and fresh top weight were made. Roots were observed for galling and multibranching before root dry weight was determined.

Results

Due to variations in experimental design and utilization of different host plants, results are summarized by host plant. Experiments utilizing carrots were analyzed with a two factor analysis of variance with temperature and cup depth as experimental variables for each soil mix. Soil moisture, soil pH and nematode population were measured responses. No significant differences were revealed in the analysis.

Cranberry seedlings and cuttings were analyzed separately with a two factor analysis of variance with temperature and nematode inoculation as experimental variables and runner growth, root dry weight and nematode population as measured responses. No significant differences were revealed in the analysis equal to or below $P = 0.05$.

Data obtained from cranberry seedlings grown in plastic petri dishes were not analyzed statistically due to extreme variability within replications and between treatments.
Effects of different temperatures and soil types on *H. similis* populations are summarized in Fig. 4. A descriptive key of measurements for pH, moisture and nematode populations are listed in Table 2. The optimum temperature range for reproduction in both soil mixes was 22°C day and 14°C night. High nematode populations corresponded to maximum root density in the base of the cups. Roots were twisted, galled and very sparse in upper levels of cups and dense multibranching was apparent in the base of cups of infested soil, however, a uniform distribution of roots was seen throughout cups in controls.

Movement of nematodes from cups of infested soil to non-infested controls was greatest with soil mix #1. Sand from Bandon was a finer texture and contained a higher percentage of clay and organic material than Newport sand. Soil mix #1 therefore, had larger pore spaces than soil mix #2 and could allow for greater nematode movement. The moisture content was highest in the lower one-third of the cup due to close contact with high free moisture in the sand in which cups were placed. A pH gradient existed in the cups at the end of the experiment. In all cases the highest value in the gradient occurred in the lower third of the cup which corresponded to maximum root density where maximum nutrient uptake should occur. In soil mix #1 the overall pH became less acid than initial measurements, however, a more acid pH occurred with soil mix #2.
Figure 4. Influence of temperature and soil type on population increase of Hemicicliophora similis on carrot.
Table 2. Soil pH, soil moisture and nematode populations from carrot seedlings grown in two soil types \(^a\) and three temperature regimes.

<table>
<thead>
<tr>
<th>Cup</th>
<th>Depth</th>
<th>Soil mix #1</th>
<th>Soil mix #2</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>pH</td>
<td>moisture</td>
</tr>
<tr>
<td></td>
<td></td>
<td>initial</td>
<td>final</td>
</tr>
<tr>
<td></td>
<td></td>
<td>30°C day/24°C night</td>
<td>22°C day/14°C night</td>
</tr>
<tr>
<td></td>
<td></td>
<td>30°C day/24°C night</td>
<td>22°C day/14°C night</td>
</tr>
</tbody>
</table>

Soil mix #1 \(^b\)

- 1: 6.2, 6.2, 14, 10.4
- 2: 6.2, 6.3, 15, 35.2
- 3: 6.2, 6.5, 18, 130.0

Soil mix #2 \(^c\)

- 1: 6.7, 6.1, 13, 4.3
- 2: 6.7, 6.4, 13, 28.0
- 3: 6.7, 6.5, 14, 179.9

\(^a\) Nematode population at the onset of the experiment was 210 nematodes/450 cc soil. Experimental time was two months.

\(^b\) These data are means of 9 replications. No significant F value was achieved due to extreme variability within replications, therefore an LSD, 0.05 was not derived.

\(^c\) These data are means of 7 replications. No significant F value was derived.

\(^d\) Cups were divided horizontally into three equal depths each containing 150 cc soil; depth 1, 2 and 3 represented the top, middle and base, respectively, of each cup.
Data obtained on germination success and plant size of remaining plants are summarized in Table 3. Germination of carrot seedlings was severely restricted by initial populations of nematodes, and relatively few carrot plants survived the duration of the experiment. This resulted in a subsequently low rate of reproduction in both soil mixes due to the lack of an adequate food source. It was suggested by Ruehle and Christie (1958) that vigorous attacks on the fibrous root system by the nematodes cause a drastic reduction in their food source which in turn limits nematode reproduction. Because of the high germination rate in controls, overcrowding restricted plant height; this was not a factor in cups of infested soil. Plants grown in infested soil mix #1 and in the two cooler temperatures grew taller than those in corresponding controls. No galling was observed in carrot roots in these experiments.

**Effects on Reproduction and Pathogenicity of Nematodes on Cranberry (Vaccinium macrocarpon Ait.)**

Data obtained from experiments utilizing cranberry cuttings to demonstrate the influence of temperature on the reproduction of *H. similis* and effects of feeding on runner growth and root dry weight are summarized in Figure 5. Optimum temperature for reproduction of *H. similis* when inoculated on to rooted cranberry cuttings was 30°C day 24°C night. This temperature was also optimum for growth of cranberry cuttings. Those plants growing in sterile bog soil had more runner growth than those in inoculated soil. This was not the case in plants grown at 22°C day 14°C night. The root dry weight of plants
Table 3. Germination percent and plant size of carrot seedlings grown in nematode infested and sterile soil under different environmental conditions. 

<table>
<thead>
<tr>
<th>Temperature Conditions</th>
<th>Mix #1</th>
<th>Mix #2</th>
<th>% Germination</th>
<th>Av. no. plant/cup</th>
<th>Av. size plant (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>30°C day 24°C night</td>
<td></td>
<td></td>
<td>Infested</td>
<td>Control</td>
<td>Infested</td>
</tr>
<tr>
<td>mix #1</td>
<td>1.0</td>
<td>7.0</td>
<td>90.0</td>
<td>45.0</td>
<td>3.0-7.0</td>
</tr>
<tr>
<td>mix #2</td>
<td>2.0</td>
<td>14.8</td>
<td>180.0</td>
<td>90.0</td>
<td>6.0-11.0</td>
</tr>
<tr>
<td>22°C day 14°C night</td>
<td></td>
<td></td>
<td>Infested</td>
<td>Control</td>
<td>Infested</td>
</tr>
<tr>
<td>mix #1</td>
<td>9.0</td>
<td>4.0</td>
<td>90.0</td>
<td>75.0</td>
<td>8.0-12.0</td>
</tr>
<tr>
<td>mix #2</td>
<td>18.3</td>
<td>8.5</td>
<td>180.0</td>
<td>150.0</td>
<td>4.0-7.0</td>
</tr>
<tr>
<td>20°C day 6°C night</td>
<td></td>
<td></td>
<td>Infested</td>
<td>Control</td>
<td>Infested</td>
</tr>
<tr>
<td>mix #1</td>
<td>12.0</td>
<td>6.0</td>
<td>90.0</td>
<td>20.0</td>
<td>2.3-7.1</td>
</tr>
<tr>
<td>mix #2</td>
<td>23.7</td>
<td>11.4</td>
<td>180.0</td>
<td>39.0</td>
<td>1.0-4.0</td>
</tr>
</tbody>
</table>

\(\text{a}\) Nematode population at the onset of the experiment was 210 nematodes/450 cc soil. Experimental time was two months.

\(\text{b}\) These data are means of 9 replications for infested soil and 3 replications for controls.

\(\text{c}\) These data are means of 7 replications for infested soil and 3 replications for controls.
Figure 3. Influence of temperature on reproduction of *Hemicycliophora similis* and feeding effects on runner growth and root dry weight of rooted cranberry cuttings (*Vaccinium macrocarpon* Ait.).
inoculated with *H. similis* grown in warmer temperatures was greater than the corresponding uninoculated plants. In cooler temperatures root weight was decreased by inoculations of *H. similis*. No galling was observed on roots of cranberry cuttings. There was no movement of nematodes through the silica sand from inoculated to non-inoculated treatments.

Data obtained from experiments which utilize cranberry seedlings to demonstrate the influence of temperature on reproduction of *H. similis* and feeding effects on runner growth and root dry weight are summarized in Figure 6. The optimum temperature for reproduction of *H. similis* on cranberry seedlings was 22°C day 14°C night. The optimum temperature for seedling runner growth was 30°C day 24°C night, similar to results obtained with cranberry cuttings. The effect of nematode inoculation on runner growth was significant at P=.10 level. Although this level of significance is well above the 0.05 level it supports a definite trend.

Cranberry seedling root growth was enhanced by inoculation of *H. similis*. These data were not supported by statistical significance, however, a trend was apparent by visual assessment and by root dry weight data. No galling was observed in roots of cranberry seedlings grown in inoculated bog soil, however, multibranching was observed as the primary effect of nematode feeding.

Experimental data for cranberry seedlings grown in plastic petri dishes in 25°C±1°C are presented in Table 4. Cranberries developed longer runners in sterile sand and soil than in inoculated
Figure 6. Influence of temperature on reproduction of Hemicycliophora similis sp. and feeding effects on runner growth and root dry weight of cranberry seedlings (Vaccinium macrocarpon Ait.)
<table>
<thead>
<tr>
<th></th>
<th>Runner growth (cm)</th>
<th>Fresh top wt (g)</th>
<th>Root dry weight (g)</th>
<th>Nematodes</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Sand</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Inoculated</td>
<td>1.22</td>
<td>0.04</td>
<td>0.01</td>
<td>163</td>
</tr>
<tr>
<td>Non-inoculated</td>
<td>1.32</td>
<td>0.04</td>
<td>0.07</td>
<td>0</td>
</tr>
<tr>
<td><strong>Soil</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Inoculated</td>
<td>5.12</td>
<td>0.07</td>
<td>0.02</td>
<td>990</td>
</tr>
<tr>
<td>Non-inoculated</td>
<td>6.72</td>
<td>0.09</td>
<td>0.01</td>
<td>0</td>
</tr>
</tbody>
</table>

*Three seedlings per container were inoculated with 150 nematodes/plant at onset of experiment; experimental time was two months; these data are means of three replications.*
treatments. Higher nematode populations developed on cranberries grown in soil than on those grown in sand. Root dry weights from plants grown in inoculated soil were greater than those from sterile controls. This effect may correspond to previous data collected from experiments with seedlings grown in larger containers of the same sterile bog soil.

Feeding effects on roots were noticeable in both sand and soil treatments (see Fig. 7 A-F). On sand, nematode feeding caused an increase in stubby roots, twisting and multibranching. Cranberries grown in sterile sand showed some stubby root characteristics but much less lateral root growth and twisting. Discoloration of roots was pronounced in plants grown in inoculated sand. Cranberries grown in inoculated bog soil had swollen multibranched roots showing severe discoloration. These roots also appeared more twisted and multibranched than roots extracted from non-inoculated checks.

Discussion

It was shown in carrot host studies that different temperature regimes effect the reproductive rate of *Hemicycliophora similis*. The nematode was found to build up a fairly high population in a short period of time which noticeably effected the normal growth of carrots. Germination of carrot seedlings was reduced by nematode feeding. The initial devastation of seedlings diminished the food
Figure 7. Roots of cranberry seedlings grown in sterile and inoculated silica sand and bog soil, A) Roots grown in sterile silica sand, B) Multibranched roots grown in inoculated silica sand, C) Stubby roots grown in inoculated silica sand, D) Roots grown in sterile bog soil, E) Twisting roots grown in inoculated bog soil, F) Swollen root tips grown in inoculated bog soil.
source substantially which adversely affected subsequent population development. At the end of the experiment plants were purplish-yellow indicating a nutritional imbalance, likely nitrogen. Due to this lack of plant vigor it is possible that normal nematode reproduction and symptom development was not expressed. No galling occurred on roots of plants in this experiment.

From the pH data recorded in Table 2, one can observe that a more acidic environment resulted with soil mix no. 2. It is possible to speculate according to De Wit et al. (1963) that the addition of Bandon sand which contained organic matter increased the organic N content in the containers. This would result in a more acidic environment. Newport sand had no organic matter and plant roots in soil mix no. 1 caused a less acidic soil. The differences in soil did not significantly influence the population increase of this nematode on carrot.

The development of a pH gradient is a well studied phenomenon. De Wit et al. (1963) explain that well nourished plants have either an external alkaline effect or an external acidic effect upon the surrounding soil media. This results from the movement of cations and anions from the soil solution into the plant roots with the exchange of H+ ions and OH- ions to maintain electroneutrality in the plant. The content of organic matter can influence the external effect the roots have on the soil by supplying additional organic N usually in the ammonical form. Roots utilize this nitrogen with a net acidic
effect due to the concomitant release of $\text{H}^+$. The uptake of nitrate nitrogen results in a less acidic soil environment due to release of $\text{HCO}_3^-$. 

Nematode populations usually grow in direct response to a healthy host plant population. Temperature in the soil environment can also directly influence activities of plant parasitic nematodes including the infection process, reproduction and mobility in the soil surrounding host tissue (Van Gundy and Rackham, 1961). In greenhouse experiments unsatisfactory results can occur from irregular distribution of nematodes in the soil media, irregular activity caused by dryness in cups and aggregations of nematodes within portions of the pot and nutritional imbalances. Seinhorst and Kozlowska (1977) found that uniformity of these factors, especially during seed germination and seedling development are important for approximating field conditions.

Aspects of the experiment and data collecting procedures could be modified in order to quantify nematode density with root density to establish that maximum nematode activity corresponds directly with maximum root growth. Such experiments with carrot were developed and started, but circumstances prevented their assay for results. Perhaps higher initial population in infested soil, longer time intervals and larger containers would have provided better data to demonstrate pathogenicity on carrot as the host.
Results of cranberry host experiments did not show dramatic differences in nematode reproductive rates at various temperatures nor were there any statistically significant pathogenic effects produced by nematodes feeding on cranberry seedlings or rooted cuttings. Possible reasons include low initial inoculum level, short growth period and a minimum number of replications in the experimental design. Changing these factors in repeat tests may elucidate statistical significance among treatments.

The trends which developed in these experiments indicate that *Hemicycliophora similis* causes a growth response in cranberry seedlings; similar growth responses did not occur when this nematode fed on rooted cranberry cuttings. Zuckerman (1961) found a consistent reduction of growth in cranberry seedlings grown in soil infested with *H. similis*. He also observed multibranching and thickening of roots grown in nematode infested soil; similar effects were noted in these experiments. Zuckerman noted cuttings grown in infested soil had smaller root systems, but the differences were not statistically significant. Results obtained from cranberry cuttings in these experiments which were established in warmer temperatures were similar to those of Zuckerman's. Dry weights of roots from inoculated cuttings, grown in 22°C day 14°C night, were lower than non-inoculated controls.

Data obtained from experiments with cranberry seedlings do not
agree with Zuckerman's findings. Nematode feeding caused an increase in seedling root dry weight in both temperature regimes. This may be due to the low population level of *H. similis* used in this experiment or that a different sub-species of *H. similis* exists which causes a different pathogenic response. However, low population levels of plant parasitic nematodes have been associated with increased root growth. In a review by Barker and Olthof (1976), under certain conditions, plant parasitic nematodes may stimulate plant growth. Low populations of *Heterodera rostochiensis*, *Heterodera tabacum*, *Meloidogyne hapla*, *Pratylenchus penetrans* and *Heterodera schachtii* may enhance growth of host plant and sometimes result in increased yields.

The results of these experiments using carrot and cranberry seedlings indicate that seed germinated in infested soil is an extremely effective model for demonstrating the effects of feeding by *H. similis*. Determinations of nematode feeding effects on tap root production would be helpful information to growers who have the nematode present in their soils and intend to grow carrots. Under optimum conditions of temperature and soil texture, large populations of this nematode build up and cause extensive damage to germination and growth of carrots. From data obtained in these experiments, it can be concluded that cranberry does not appear to be a host which supports substantial reproduction of *H. similis*. 
However, the use of an infested soil source and higher inoculum levels would likely have produced a more noticeable growth response.
SECTION III: LIFE HISTORY AND FEEDING OBSERVATIONS OF *Hemicycliophora similis* THORNE, 1955

Preface

Life cycle information of plant parasitic nematodes is requisite for effective pest control programs when these pests endanger yields and quality of economic crops. Data obtained from preliminary host range studies indicate that *Hemicycliophora* reproduced and formed galls on carrot seedlings which was the major host used for this study.

The object of this investigation was to determine the number of days required to complete a life cycle and the various stages as the nematode developed from egg to egg. Data obtained from this study along with other information derived from this research will aid in a more complete characterization of this species.

Materials and Methods

To determine the number of days between each larval stage, nematode inoculum was obtained from a stock greenhouse culture, processed, separated into individual larval stages and surface sterilized. Ten to twenty nematodes of similar stages were inoculated onto carrot seedlings in 0.5% water agar plates.

Modifying the procedure outlined by Van Gundy (1959), gravid
females were placed in fresh tap water on a depression slide and stored in foil covered trays. To insure adequate oxygen and reduce fungal contamination, water was changed every other day by means of an automatic pipet. After emerging from eggs, second-stage larvae were placed in clean depression slides filled with fresh tap water. The chlorine solution used for surface sterilization in previous inoculations was too harsh for second-stage larvae so before inoculation larvae were removed to sterile distilled water in clean depression slides. To reduce contamination, sterile distilled water was changed three times before larvae were inoculated onto carrot seedlings grown in 0.5% water agar. Cultures were stored in opaque covered vegetable crispers at 25°C±2°C and checked daily to note the presence of eggs, molting nematodes and appearance of subsequent stages.

To concurrently follow the life cycle in a soil environment, carrots were planted in cups containing 150 cc sterile greenhouse soil mix and inoculated with 100 hand picked second-stage larvae. Cup contents were processed by placing all soil and roots directly onto funnel apparatus and the processing water was drained every three days to search for subsequent stages.

Results

A diagrammatic life cycle of *H. similis* tracing the number of
days from egg to adult appears in Figure 8. The accumulated time in days for developmental stages to appear in water agar cultures and soil inoculations is listed in Table 5.

**Egg**

Gravid females deposited from one to many eggs within 24 hours after being placed in depression slides filled with tap water. Usually eggs were deposited singly by the female and adhered tightly to the glass slide by means of a sticky substance associated with the outer membrane. Newly layed eggs contained one to two cells and occasionally four averaging 32x91 microns in size. Six hours after deposition of a two-celled egg, development had proceeded to the four-celled stage. Within eight hours the 8-celled stage was reached and after eighteen hours the egg contained many cells and exact numbers were difficult to distinguish.

**First-Stage Larvae**

First-stage larvae moved within the egg three days after deposition. After three to five days a transparent region in the head and neck was observed to contain cuticular globules indicating a stylet primordium (see Fig. 8). Dark vacuolation in the middle body region indicated the presence of intestinal primordia; larvae did not possess
Figure 8. A diagrammatic life cycle of *Hemicycliophora similis* Thorne, 1955.
Table 5. The number of days occurring between egg deposition and appearance of adults of *Hemicycliophora similis* as determined in greenhouse and laboratory investigations.

<table>
<thead>
<tr>
<th>Stage</th>
<th>Accumulated time and appearance of developmental stage in water agar cultures</th>
<th>Accumulated time and appearance of developmental stage in greenhouse inoculations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Egg</td>
<td>appearance = day 1</td>
<td></td>
</tr>
<tr>
<td>First-stage larvae</td>
<td>3-5 days</td>
<td></td>
</tr>
<tr>
<td>First molt</td>
<td>6-7 days</td>
<td></td>
</tr>
<tr>
<td>Second-stage larvae</td>
<td>8-10 days</td>
<td></td>
</tr>
<tr>
<td>Second molt</td>
<td>20-22 days</td>
<td></td>
</tr>
<tr>
<td>Third-stage larvae</td>
<td>23-25 days</td>
<td></td>
</tr>
<tr>
<td>Third-molt</td>
<td>30-32 days</td>
<td></td>
</tr>
<tr>
<td>Fourth-stage larvae</td>
<td>33-35 days</td>
<td></td>
</tr>
<tr>
<td>Fourth molt</td>
<td>40-42 days</td>
<td></td>
</tr>
<tr>
<td>Adult</td>
<td>43-45 days</td>
<td></td>
</tr>
<tr>
<td>Egg</td>
<td>45-47 days</td>
<td></td>
</tr>
</tbody>
</table>
a sheath or stylet at this stage. The first molt occurred six to seven days after deposition with sheath and spear clearly visible within the egg.

Second-stage larvae hatched in 8-10 days with a loose fitting sheath attached at the lips and just above the tip of the tail. Larvae maintained a stiff posture for approximately one hour after hatching and activity resumed shortly afterward.

**Second-Stage Larvae**

Second-stage larvae required a feeding period to successfully develop into subsequent stages. This was noted when larvae were found dead in depression slide cultures if left over one week. After inoculation onto carrot seedlings, second-stage larvae underwent a lengthy exploratory period which involved probing at root tips and moving in and around several root tips before selecting a feeding site. This time period lasted up to 10-15 days before larvae began to feed. Feeding larvae usually oriented themselves just behind the root tip with stylets inserted several cells deep and remained for one to four days. Sometimes they withdrew their stylets and moved to another root tip and resumed feeding.

Feeding terminated as nematodes began to molt and remained inactive in a half-moon position for one to three days. After the quiescent period, vigorous activity ensued resulting in the splitting
of the old larval cuticle, wriggling free and searching for a new feeding site. Larvae undergoing the second molt were found in soil wash water extracted from carrot seedlings 4-6 days after inoculation of second-stage larvae.

**Third-Stage Larvae**

Third-stage larvae were first observed 12-15 days after second-stage larvae were inoculated onto carrot seedlings. These larvae were sparsely vacuolated with loosely fitting sheaths. Third-stage larvae were distinct from second-stage larvae due to increased size, tail shape and length of stylet and esophagus. They required an exploratory-feeding period of 10-13 days, as observed in water agar cultures, before successfully molting to a fourth-stage larva. Larvae undergoing the third molt were extracted 21-23 days after initial inoculation and 9 days after the first indication of third-stage larvae. They were dark throughout the body and had tight fitting sheaths.

**Fourth-Stage Larvae**

In soil cultures fourth-stage larvae were extracted 24-26 days after initial inoculations, 13 days from the first indication of third-stage larvae. Information obtained from water agar cultures corresponded agreeably with soil cultures. Third-stage larvae molted
to fourth-stage larvae in 10-12 days when inoculated onto carrot
seedlings grown in water agar. Fourth-stage larvae were distinct
from third-stage larvae in tail shape, increased body length and
length of the stylet and esophagus. Larvae undergoing a fourth molt
were observed 31-34 days after initial inoculations of second-stage
larvae on to carrot seedlings in soil cultures.

Fourth-stage larvae required a less lengthy exploration time
before feeding and moving from root to root; feeding time varied
from a few hours to three days. Feeding and development time
between fourth-stage larvae and young adults was 10-12 days.
Some of these nematodes remained alive for three to four weeks
in water agar cultures without molting.

**Adult Females**

Adult females were extracted from soil cultures 35-38 days
after initial inoculation with second-stage larvae. Eggs were seen
soon afterward in soil wash water.

Having completed the final larval molt, young adults that
were removed from water agar cultures and inoculated onto new
agar cultures, began feeding within 24 hours. As soon as three days
after feeding resumed, gravid females deposited eggs which subse-
quently hatched in 8-10 days. Some adults remained alive in water
agar for four to five weeks at 23°C±2°C without feeding; these
nematodes did not lay eggs. The total time elapsed from egg to egg was 45-47 days at 23°C±2°C on host carrot seedlings grown in water agar cultures.

Discussion

Van Gundy (1959) suggests that both the disease cycle and life cycle of this group of ectoparasitic nematodes is influenced more by the soil environment than are the endoparasites and sedentary ectoparasitic nematodes. It has not been determined how this nematode behaves in its natural environment since field observations were not undertaken. The life cycle of this ectoparasitic nematode was observed and developed in an unnatural environment by combining laboratory and greenhouse methods. Laboratory derived data pertaining to life histories, pathogenic capabilities and taxonomic characteristics are helpful in characterizing the species. Since similar results were achieved in both sterile soil and water agar cultures the credibility of these methods and results is maintained.

Many biotic and abiotic factors effect the reproduction of nematodes. In water agar cultures many of these factors were absent, such as soil temperature fluctuations, moisture stress and predatory microorganisms. The nematodes inoculated onto the carrot seedling cultures did not encounter competition for the host from any other nematodes, sporozoans, bacteria or fungi. Primary problems also
existed with the development and physiology of the host. The nutrition of the host plant affects the rate at which nematodes develop according to Oteifa (1963) and Bird (1960). Mountain (1965) relates that the rate of reproduction of nematodes tends to be highest when host plants receive full nutritional requirements. In water agar, full root development is restricted by many artificial conditions including lack of an adequate supply of nutrients. Sterile soil was used in greenhouse life cycle studies to insure freedom from competition by other plant pathogenic nematodes, bacteria and fungi. Carrot plants probably obtained a better nutrient supply and growing conditions when grown in a soil medium.

Environmental factors such as an oxygen deficiency may have influenced the molting process of nematodes in water agar cultures. A molting nematode may be at one of the weakest points in its life cycle for withstanding stress (Van Gundy, 1965). Body tissues have a high metabolic need for oxygen when nematodes are producing a new cuticle and other structures including a new stylet in plant parasitic forms. Oxygen deficiency may have been a factor for the low numbers of nematodes which successfully molted to the subsequent stage.

An interesting observation made during microscopic examinations of H. similis populations was the appearance of embryonating eggs within the fourth-stage larval cuticle (see Fig. 9). This
Unusual occurrence of embryonating eggs lodged within the fourth larval and adult cuticle.

A) Two embryonating eggs within fourth larval cuticle of a molting fourth-stage larvae,

B) Two-celled egg within the fourth larval cuticle,

C) Egg within shed cuticle of a fourth-stage larvae,

D) Embryonating egg within adult cuticle.
phenomenon was observed in nematodes in extraction water from carrot, cranberry and peppermint and with those gravid females placed in depression slides for life history investigations.

The specimen in Fig. 9A is undergoing the last larval molt (note the absence of a stylet and the presence of three cuticles in the tail region) and contains two embryonating eggs. The eggs are lodged within the fourth larval cuticle. In Fig. 9B a two-celled egg is lodged within the fourth-stage larval cuticle which is still attached to the adult female. By noting the tail shape of the fourth-stage larvae in Fig. 10C it is apparent that the specimen in Fig. 9C shows an egg within a shed cuticle of a fourth-stage larvae. The adult female pictured in Fig. 9D has an embryonating egg lodged within the normal adult sheath; as many as seven eggs in all stages of development were observed lodged within the unshed fourth larval cuticle. Specimens became contaminated with fungi before pictures could be taken. Eggs were seen to have developed through second-stage larvae, however, hatching within the cuticle was not observed.

It is difficult to determine the exact cause of this phenomenon. It appears that eggs are lodged within the cuticle because of an incomplete molt, possibly the remnants of the fourth larval cuticle prevent normal egg deposition. Normally the old cuticle and sheath degenerates and is either reabsorbed or simply broken down (Johnson et al., 1970). Frequent observations of remnants of clinging old sheath and cuticle...
indicate normal molting processes are being impeded, either by a stressful environment, nutritional or oxygen deficiencies or possibly disease. Much additional information is required to provide a more complete understanding of this phenomenon.
SECTION IV: TAXONOMY (MORPHOMETRICS) OF *Hemicycliophora similis*

Preface

Deriving the exact species name and identity of a plant parasitic nematode aids the taxonomist, biologist and plant pathologist in cataloging information obtained from research studies. Accurate identification of a plant parasitic nematode is often subjective on the part of taxonomists since differences of opinion may occur. The exact species identity of this nematode is still controversial. The key of species in the genus *Hemicycliophora* proposed by Thorne (1955) separates *H. similis* and *H. typica* by the lack of males associated with *H. similis* and their presence associated with *H. typica*. No males have been observed in this study. Morphometric investigations and communications with taxonomists (nematologists and professors in nematology) were utilized for this taxonomic study. For purposes of this research the nematode studied was identified as *Hemicycliophora similis* Thorne, 1955.

Materials and Methods

Fresh mounts of specimens were prepared by placing nematodes collected from soil wash water in a drop of water which was first heated to relax nematodes. Glass rods were inserted to
prevent the nematodes from being flattened by the weight of the cover slip. Specimens were then covered with a No. 1 glass cover slip and sealed with clear fingernail polish. Measurements were made of all stages of development. The negatives were later placed in a photographic enlarger and drawn to a scale of 2000x magnification.

**Results and Discussion**

Measurements of this nematode determined by microscopic investigations are compared with measurements reported by Thorne (1955) and Brzeski (1974) in Table 6; a descriptive key of measurement symbols is listed in Table 7. Thorne's key includes 15 species; Brzeski expanded the key to 69 species. Besides those measurements of Thorne's listed in Table 6, he also included the location of the vulva to be 48-66 annules from the tail; anus exceedingly obscure about 20 annules behind the vulva and the tail is convex-conoid. The Rva for the specimen from Bandon is 26.9, this varies greatly from Thorne's measurement of 48-66. The Rva reported by Brzeski, 20(15-23) is more closely related to the Bandon specimen.

Taxonomic characteristics of *H. similis* in Brzeski's key did not coincide closely with stylet measurements, vulva and tail descriptions and R values of this nematode. The majority of values listed by Brzeski are at the low end of variations noted with the Bandon specimen. The general variation of most measurements prompted
Table 6. Measurements of adult *Hemiycycliphora similis* found in Bandon, Oregon compared with those derived by Thorne (1955) and Brzeski (1974).

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>L</td>
<td>1.24 mm (1.18-1.33)</td>
<td>1.1 mm</td>
<td>1.16 mm (1.11-1.20)</td>
</tr>
<tr>
<td>W</td>
<td>40.3 u (37.6-43.2)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>a</td>
<td>30.9 (28.1-33.6)</td>
<td>23</td>
<td>-</td>
</tr>
<tr>
<td>b</td>
<td>6.4 (6.1-6.9)</td>
<td>5.6</td>
<td>6.2 (5.2-6.8)</td>
</tr>
<tr>
<td>c</td>
<td>8.6 (8.2-9.0)</td>
<td>-</td>
<td>10.8 (9.9-11.3)</td>
</tr>
<tr>
<td>S+</td>
<td>101.5 u (98.4-107.2)</td>
<td>[102.25 (81.07-123.42)]</td>
<td>92 u (88-96)</td>
</tr>
<tr>
<td>esoph</td>
<td>193.7 u (186.4-199.2)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>tail</td>
<td>146.0 u (135.2-160.0)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>V</td>
<td>82 u (80-83)</td>
<td>51-79</td>
<td>85 (84-87)</td>
</tr>
<tr>
<td>R</td>
<td>324.8 (306.0-343.0)</td>
<td>290-307</td>
<td>293 (276-305)</td>
</tr>
<tr>
<td>Rex</td>
<td>56.6 (53-61)</td>
<td>-</td>
<td>54 (49-56)</td>
</tr>
<tr>
<td>Rv</td>
<td>243.4 (236-253)</td>
<td>-</td>
<td>240 (223-251)</td>
</tr>
<tr>
<td>Rva</td>
<td>26.9 (23-31)</td>
<td>-</td>
<td>20 (15-23)</td>
</tr>
<tr>
<td>Rt</td>
<td>52.5 (44-59)</td>
<td>-</td>
<td>33 (30-40)</td>
</tr>
<tr>
<td>VT/VB</td>
<td>7.6 (7.2-7.9)</td>
<td>-</td>
<td>4.5 (4.1-5.2)</td>
</tr>
</tbody>
</table>

*d* Thorne reported the stylet extended through 22-34 annules. This figure was derived by multiplying the length of one annule or Thorne's specimen times the number of annules that the stylet extended.
Table 7. Descriptive key to symbols used in measurements of *Hemicycliophora similis*.

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>L</td>
<td>body length in millimeters,</td>
</tr>
<tr>
<td>W</td>
<td>greatest body width (usually at vulva),</td>
</tr>
<tr>
<td>a</td>
<td>ratio of body length to body width at the end of the pharynx,</td>
</tr>
<tr>
<td>b</td>
<td>ratio of body length to the length of the pharynx,</td>
</tr>
<tr>
<td>c</td>
<td>ratio of body length to the length of the tail,</td>
</tr>
<tr>
<td>St</td>
<td>stylet length in microns,</td>
</tr>
<tr>
<td>esoph</td>
<td>esophagus length in microns (tip of stylet to base of esophagus),</td>
</tr>
<tr>
<td>tail</td>
<td>length of tail in microns (anus to tip of tail),</td>
</tr>
<tr>
<td>V</td>
<td>position of the vulva measured from the anterior end of the body, as a percentage of the total body length,</td>
</tr>
<tr>
<td>R</td>
<td>number of body annules on the ventral side,</td>
</tr>
<tr>
<td>Rex</td>
<td>number of annules from the anterior end of the body to the first annules posterior to the excretory pore,</td>
</tr>
<tr>
<td>Rv</td>
<td>number of annules from the anterior end of the body to the vulva,</td>
</tr>
<tr>
<td>Pva</td>
<td>number of annules from the vulva to the first annule posterior to the anus,</td>
</tr>
<tr>
<td>Rt</td>
<td>number of annules on the tail,</td>
</tr>
<tr>
<td>VT/VB</td>
<td>ratio of the post-vulval body length to body width immediately posterior to the vulva.</td>
</tr>
</tbody>
</table>
Golden (personal communication) to suggest the creation of a new species. Esser (personal communication) did not find enough difference to establish a new species; consultation with Jensen resulted in a similar prognosis. Further communications with Zuckerman indicated a similarity of this nematode to the H. similis found in Massachusetts cranberry bogs.

Conflicts noted between measurements recorded by Brzeski for stylet length, annulation and VT/VB values were not considered by Esser to be significant enough to warrant a new species. He explained that since Thorne used a single specimen to describe the species there is no population variation allowed for in the original description. His morphometrics are subject to + and - variations thus specific dimensions can be rationally resolved.

The drawings in Fig. 10 follow the development of this nematode through successive larval stages. A list of measurements of all developmental stages is found in Table 8, the measurement symbols are the same as those explained in Table 7. The excretory pore can be seen in Figs. 10H to open 5 annules below the base of the esophagus. This same structure is pictured by Brzeski to open two annules below the esophagus, and three annules below the esophagus in Thorne's specimen.

A soil sample containing the nematode was received by the Oregon State University Plant Clinic in October 1972. Since that time
the nematode was maintained on peppermint (Mentha piperita var. 'Todd' and 'Mitcham') in the nematology greenhouse. During that time it is possible that selective pressures have resulted in morphological variations within the population. This could account for the variation in measurements that have been observed.

Variability of numerous taxonomic characteristics are reported in nematology literature. In morphological observations of Rotylenchus goodeyi, Coomans (1962) found variations in body dimensions, head annulation, spear knobs, esophagus length, female gonad length and tail length. Taylor and Jenkins (1957) found variations in tail shape and size in naturally occurring Pratylenchus populations. Tail shape variability has been observed in Hemicycliophora zuckermani by Brzeski and Zuckerman (1965) and by Minton and Golden (1956, 1966). Because research has found such variability in morphology within nematode populations further characterization of this species by biology and pathogenicity studies seems a necessity.
Figure 10. Scale drawings of heads and tails of all developmental stages of Hemicycliophora similis, A) head of second-stage larvae, B) head of third-stage larvae, C) head of fourth-stage larvae, D) head of adult female, E) tail of second-stage larvae, F) tail of third-stage larvae, G) tail of fourth-stage larvae, H) tail of adult female.
Table 8. Measurements of *Hemicycliophora similis* native to cranberry bogs in Bandon, Oregon.

<table>
<thead>
<tr>
<th></th>
<th>ADULT</th>
<th>IV</th>
<th>III</th>
<th>II</th>
</tr>
</thead>
<tbody>
<tr>
<td>L</td>
<td>1.25 mm (1.18-1.33)</td>
<td>.853 mm (.772-.998)</td>
<td>.619 mm (.552-.661)</td>
<td>.426 mm (.379-.470)</td>
</tr>
<tr>
<td>W</td>
<td>40.3 μ (37.6-43.2)</td>
<td>25.6 μ (24.0-32.8)</td>
<td>20.9 μ (19.2-22.4)</td>
<td>18.7 μ (15.2-23.2)</td>
</tr>
<tr>
<td>a</td>
<td>30.9 (28.1-33.6)</td>
<td>31.0 (29.6-33.0)</td>
<td>29.7 (28.2-31.1)</td>
<td>22.9 (20.9-25.9)</td>
</tr>
<tr>
<td>b</td>
<td>6.4 (6.1-6.9)</td>
<td>5.2 (4.7-6.2)</td>
<td>4.5 (4.3-4.9)</td>
<td>3.7 (3.4-4.7)</td>
</tr>
<tr>
<td>c</td>
<td>8.6 (8.2-9.0)</td>
<td>7.8 (6.9-8.5)</td>
<td>7.6 (7.2-8.3)</td>
<td>7.4 (6.6-8.2)</td>
</tr>
<tr>
<td>St</td>
<td>101.5 μ (8.4-107.2)</td>
<td>87.9 μ (82.4-92.0)</td>
<td>73.0 μ (67.2-76.8)</td>
<td>56.6 μ (50.4-60.0)</td>
</tr>
<tr>
<td>esoph</td>
<td>193.7 μ (186.4-199.2)</td>
<td>162.6 μ (155.2-171.2)</td>
<td>138.1 μ (128.8-145.6)</td>
<td>114.1 μ (107.2-123.2)</td>
</tr>
<tr>
<td>tail</td>
<td>146.0 μ (135.2-160.0)</td>
<td>110.0 μ (102.4-119.2)</td>
<td>81.6 μ (72.8-86.4)</td>
<td>58.1 μ (52.8-69.6)</td>
</tr>
<tr>
<td>V</td>
<td>82 μ (80-83)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>R</td>
<td>324.8 (306.0-343.0)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rex</td>
<td>56.6 (53-61)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rv</td>
<td>243.4 (236-253)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rva</td>
<td>26.9 (23-31)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rt</td>
<td>52.5 (44-59)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>VT/VB</td>
<td>7.6 (7.2-7.9)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
CONCLUSIONS

Greenhouse and laboratory studies were made of *Hemicyclophora similis* found in cranberry bogs in Bandon, Oregon to determine preferred hosts, to examine optimum environmental conditions for reproduction and pathogenic expression, and to obtain information on its life history.

Results of host range studies indicated that *H. similis* can reproduce and form galls on beet, carrot, rutabaga and turnip; and multibranched growths on a variety of plant species including broccoli, dill, and radish; galling was most severe on carrot. Corn, pea, both radish varieties and rutabaga supported nematode populations which developed over a 100% increase from initial inoculum. Beet, broccoli, corn and dill were designated as non-hosts since populations reduced, remained static or did not increase over 100% of initial inoculum. Germination of carrot seedlings was severely reduced when grown in sandy soil infested with *H. similis*. Less substantial reductions in germination occurred with beet, dill, radish, rutabaga, tomato and turnip. Carrot, tomato and turnip supported the highest populations of the nematode.

Cranberry cuttings and seedlings and carrot seedlings were used as hosts to study the influence of environmental factors such as temperature and soil pH and texture on reproduction and pathogenicity.
Carrot seedlings supported the highest populations in both soils tested at 22°C day and 14°C night. Dense multibranching and twisting of roots was observed in plants grown in nematode infested soil. Maximum root densities and nematode populations were found at the base of cups. A pH gradient developed in cups containing carrot seedlings; a more acid condition developed in the mix containing Bandon, Oregon bog sand and a more alkaline condition developed in the soil mix containing Newport, Oregon sand. The gradient developed in response to nutrient uptake by plants and the simultaneous release of ions to maintain electroneutrality within plant roots. Nematode populations on cranberry cuttings were highest at 30°C day and 24°C night. Growth of cranberry cuttings was not effected by nematode feeding in either temperature, however, runner growth and root dry weights of inoculated cranberry seedlings were greater than in non-inoculated controls in both temperatures. Roots of inoculated cranberry seedlings were twisted and multibranched; roots of cuttings did not show any abnormalities.

The life cycle of *H. similis* was determined by using carrot seedlings as a host in laboratory and greenhouse studies. The complete life cycle lasts 45-47 days at 23°C±2°C as the nematode developed through successive larval stages from egg to egg.

For purposes of this research this nematode was called *Hemicycliophora similis*. Morphometric studies revealed deviations
from measurements recorded by Thorne (1955) and Brzeski (1974) for *H. similis*. Personal communications with R. P. Esser, H. J. Jensen and B. M. Zuckerman indicated that these slight variations did not necessitate the creation of a new species. Embryonating eggs were observed to be lodged within an incompletely shed fourth larval cuticle and adult cuticle. In some cases these eggs developed through second-stage larvae, but none was observed to emerge free of the adult.

Results of the completed experiments give a reasonably complete characterization of the biology, pathogenicity and life history of this nematode. Information derived from this research does not provide conclusive evidence that *H. similis* is a destructive pest to cranberry production. Further research exploring the response of increased root growth of cranberry seedlings to inoculations of *H. similis* may reveal a beneficial relationship between nematode and enhanced plant growth.
LITERATURE CITED


Taylor, D. P. and W. R. Jenkins. 1957. Variation within the nematode genus *Pratylenchus* with descriptions of *P. hexincisus* n sp. and *P. subpenetrans* n sp. Nematologica 2:159-179.


advances with emphasis on plant parasitic and soil forms. Univ. N. Carolina Press, Chapel Hill, N. C.


APPENDICES
APPENDIX A

Fertilizer Mixtures
APPENDIX A

Urea-Formaldehyde Urea Mixture

Incorporation of 0.34 g/l of soil of the mixture occurred prior to planting cranberry cuttings and seedlings to provide an adequate supply of essential nutrients.

<table>
<thead>
<tr>
<th>Composition</th>
<th>% N</th>
<th>% P</th>
<th>% K</th>
<th>grams</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urea-formaldehyde</td>
<td>38</td>
<td>0</td>
<td>0</td>
<td>132</td>
</tr>
<tr>
<td>Urea</td>
<td>45</td>
<td>0</td>
<td>0</td>
<td>112</td>
</tr>
<tr>
<td>Super phosphate</td>
<td>20</td>
<td>20</td>
<td>0</td>
<td>500</td>
</tr>
<tr>
<td>Muriate of potash</td>
<td>10</td>
<td>40</td>
<td>60</td>
<td>167</td>
</tr>
<tr>
<td>Hydrated lime</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>20</td>
</tr>
<tr>
<td>Dolomite</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>69</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td></td>
<td></td>
<td>1000</td>
</tr>
</tbody>
</table>

Hoagland's Solution

Cranberry seedlings planted in plastic petri dishes were watered with this solution.

<table>
<thead>
<tr>
<th>Stock 1</th>
<th>g/l</th>
<th>Stock 2</th>
<th>g/l</th>
<th>The complete solution contained:</th>
<th>ml/l</th>
</tr>
</thead>
<tbody>
<tr>
<td>KH₂PO₄</td>
<td>13.5</td>
<td></td>
<td></td>
<td>Stock 1</td>
<td>5.0</td>
</tr>
<tr>
<td>KNO₃</td>
<td>50.5</td>
<td></td>
<td></td>
<td>Stock 2</td>
<td>5.0</td>
</tr>
<tr>
<td>MgSO₄</td>
<td>24.0</td>
<td></td>
<td></td>
<td>Iron Solution</td>
<td>5.0</td>
</tr>
<tr>
<td>(Trace Elements)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>g/l</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>H₂BO₃</td>
<td>1.82</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MnSO₄·H₂O</td>
<td>2.10</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ZnSO₄·7H₂O</td>
<td>0.656</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CuSO₄·5H₂O</td>
<td>0.20</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MoO₃</td>
<td>0.034</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stock 2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ca(NO₃)₂·4H₂O</td>
<td>118.0</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stock 2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FeSO₄·7H₂O</td>
<td>2.4</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>EDTA Na₂</td>
<td>2.8</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Somogyi et al. (1964).*