Copyright Statement

The digital copy of this thesis is protected by the Copyright Act 1994 (New Zealand).

This thesis may be consulted by you, provided you comply with the provisions of the Act and the following conditions of use:

- you will use the copy only for the purposes of research or private study
- you will recognise the author's right to be identified as the author of the thesis and due acknowledgement will be made to the author where appropriate
- you will obtain the author's permission before publishing any material from the thesis.
AN INVESTIGATION OF THE SUSCEPTIBILITY OF VARIETIES
OF LUCERNE MEDICAGO SATIVA L. TO THE NEMATODE
DITYLENCHUS DIPSACI (KÜHN)

A thesis
submitted in partial fulfilment
of the requirements for the Degree
of
Master of Agricultural Science
in the
University of Canterbury

by

P.A. Burnett

Lincoln College

1971
FRONTISPIECE

Field of lucerne in Mid Canterbury

Lighter areas indicate nematode damage
<table>
<thead>
<tr>
<th>CHAPTER</th>
<th>CONTENTS</th>
</tr>
</thead>
<tbody>
<tr>
<td>I.</td>
<td>INTRODUCTION ........................................... 1</td>
</tr>
<tr>
<td>II.</td>
<td>REVIEW OF THE LITERATURE ................................. 3</td>
</tr>
<tr>
<td></td>
<td>1. Taxonomy ................................................. 3</td>
</tr>
<tr>
<td></td>
<td>2. Host Range .............................................. 4</td>
</tr>
<tr>
<td></td>
<td>3. Biological Races ........................................ 6</td>
</tr>
<tr>
<td></td>
<td>4. Life History .............................................. 8</td>
</tr>
<tr>
<td></td>
<td>5. Resistance Testing Techniques ......................... 11</td>
</tr>
<tr>
<td>III.</td>
<td>MATERIALS AND METHODS ....................................... 19</td>
</tr>
<tr>
<td></td>
<td>1. Nematode Collection, Preservation and Extraction .... 19</td>
</tr>
<tr>
<td></td>
<td>2. Plant Preparation ........................................ 25</td>
</tr>
<tr>
<td></td>
<td>3. Plant Inoculation ........................................ 29</td>
</tr>
<tr>
<td>IV.</td>
<td>RESULTS ..................................................... 35</td>
</tr>
<tr>
<td></td>
<td>1. Results from soil flats trials ......................... 35</td>
</tr>
<tr>
<td></td>
<td>2. Results from filter paper rolls ....................... 38</td>
</tr>
<tr>
<td>V.</td>
<td>DISCUSSION AND CONCLUSION ................................ 64</td>
</tr>
<tr>
<td>VI.</td>
<td>SUMMARY .................................................. 69</td>
</tr>
<tr>
<td>ACKNOWLEDGEMENTS</td>
<td>........................................... 70</td>
</tr>
<tr>
<td>REFERENCES</td>
<td>................................................ 71</td>
</tr>
<tr>
<td>APPENDICES</td>
<td>.................................................. 75</td>
</tr>
<tr>
<td></td>
<td>Appendix 1 .................................................. 75</td>
</tr>
<tr>
<td></td>
<td>Appendix 2 .................................................. 84</td>
</tr>
</tbody>
</table>
# LIST OF FIGURES

<table>
<thead>
<tr>
<th>FIGURE</th>
<th>PAGE</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. An outline of the life history of Ditylenchus dipsaci</td>
<td>10</td>
</tr>
<tr>
<td>2. Apparatus for the extraction of nematodes from plant material</td>
<td>21</td>
</tr>
<tr>
<td>3. Apparatus for separating nematodes from soil and plant debris</td>
<td>23</td>
</tr>
<tr>
<td>4. Apparatus for separating nematodes from soil and plant debris</td>
<td>24</td>
</tr>
</tbody>
</table>
## LIST OF PLATES

<table>
<thead>
<tr>
<th>PLATE</th>
<th>Description</th>
<th>PAGE</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Covered trays used to hold the filter paper rolls in the growth cabinet</td>
<td>27</td>
</tr>
<tr>
<td>2.</td>
<td>Syringe used for inoculation of lucerne seedlings</td>
<td>30</td>
</tr>
<tr>
<td>3.</td>
<td>Lucerne plants in filter paper rolls three weeks after inoculation</td>
<td>32</td>
</tr>
<tr>
<td>4.</td>
<td>Nematode infected and uninfected plants after three weeks</td>
<td>33</td>
</tr>
</tbody>
</table>

**FRONTISPIECE**

Field of lucerne in Mid Canterbury.
LIST OF TABLES

TABLE PAGE
1. Condition of young lucerne plants (Wairau) two weeks after infection with nematodes in 1% solution of methyl cellulose .......................... 36
2. Condition of young lucerne plants (Wairau) three weeks after infection with nematodes in 1% solution of methyl cellulose ..................... 37
3. Condition of young lucerne plants (Wairau) four weeks after infection with nematodes in 1% solution of methyl cellulose ..................... 37
4. Results from filter paper rolls in Experiment 1 .................................. 39
5. Results from rolls inoculated with nematodes in 1% solution of methyl cellulose ......................................................... 41
6. Mortalities in rolls inoculated with 1% solution of methyl cellulose ................. 42
7. Mortalities in control rolls .................................................................... 43
8. Results from three lucerne varieties tested for nematode susceptibility ......................................................... 44
9. Results from three lucerne varieties tested for nematode susceptibility ................. 47
10. Results from the macerated seedlings from Experiment 11 ......................... 49
11. Results from three lucerne varieties tested for nematode susceptibility ..................... 51
12. Results from the macerated seedlings from Experiment 12 ......................... 53
LIST OF TABLES
(Continued)

TABLE

13. Results from three lucerne varieties tested for nematode susceptibility

14. Results from the macerated lucerne seedlings from Experiment 13

15. Mean stem diameters in mm. of 45 seedlings of three varieties of lucerne in three experiments

16. Transformed data from Table 15 - (log_{10} of mean stem diameter)

17. Analysis of variance of results in Table 16

18. Analysis of variance of plants containing nematodes and eggs

19. Percentage of plants containing reproducing nematodes

20. Comparison of stem diameter of seedlings with ratings of nematode numbers and reproduction within the plants for 30 plants from Experiment 13

PAGE

55
57
58
59
60
61
61
63
INTRODUCTION

The stem nematode, *Ditylenchus dipsaci* (Kühn), is an important pest of lucerne (*Medicago sativa* L.), in Canterbury. Because of the way in which lucerne is grown, the nature of the crop and its monetary value, the introduction or development of varieties resistant to nematode attack seems to be the most promising method of combating this pest at present.

In other parts of the world, nematodes similar to *Ditylenchus dipsaci* have been successfully controlled on some crops by the use of systemic organo-phosphate insecticides. However, the cost of these chemicals usually exceeds the value of the lucerne crop, a factor which also excludes the use of soil fumigants.

Crop rotation with non host plants also affords an economical method of controlling *Ditylenchus dipsaci*. However, frequent crop rotation is not favoured in Canterbury as the cost of lucerne establishment is high and, on the lighter lands, profitable non host plants are difficult to find.

Control of nematodes by the introduction or maintenance of their parasites, predators or diseases has been tried in other countries several times. When it has been possible to alter the soil environment to favour parasites or predators, control has been occasionally achieved. However, just what conditions are necessary for the successful propagation of the enemies of *Ditylenchus dipsaci* are unknown at present.

Several nematode resistant strains of lucerne have been developed in North and South America. Unfortunately, none of these are agronomically
suited to New Zealand conditions. Further, just how susceptible the major New Zealand variety of lucerne, Wairau, is to nematode attack in comparison to the resistant varieties developed overseas, has not been determined. It was decided, therefore, to develop a technique to test the ability of various varieties of lucerne to withstand the attacks of the Canterbury race of *Ditylenchus dipsaci*. Because of the large host range and the many different races of *Ditylenchus dipsaci* reported in literature, it was deemed important to obtain nematodes belonging to the particular race attacking Canterbury grown lucerne.
CHAPTER II

REVIEW OF THE LITERATURE

1. TAXONOMY

The major stem nematode disease of lucerne, Medicago sativa L. is caused by the obligate endoparasitic nematode, Ditylenchus dipsaci (Kuhn). Its classification after Thorne (1949) and Goodey (1963) is as follows:

- Phylum: Aschelminthes
- Class: Nematoda
- Order: Tylenchida
- Superfamily: Tylenchoidea
- Family: Tylenchidae
- Subfamily: Tylenchinae
- Genus: Ditylenchus Filipjev, 1936

Type species Ditylenchus dipsaci (Kuhn, 1857) Filipjev, 1936.

Thorne (1949) states that this nematode ranges in length from 1.0 mm. to 1.3 mm. and gives a detailed description of the species. The nematode was first described by Kuhn as Anguilla dipsaci from specimens taken in Fuller's teasel, Dipsacus fullonum L. (Bingefors, 1957, and Barker and Sasser, 1959). Due to the large host range of this species many synonyms were described in early literature. Thorne (1945) and Goodey (1963) include synonym lists.
2. HOST RANGE

This stem infecting nematode is believed to have been present in Germany at least as early as 1919, when Schweiz (Edwards, 1932) described a disease of red clover very similar to the condition now known to be caused by Ditylenchus dipsaci. The first record of the occurrence of Ditylenchus dipsaci on lucerne was made by Kuhn in 1931 (Edwards, 1932), who described it as a new species Tylenchus navensteinii. Later work by Ritzema Bos (Edwards, 1932) showed this form was in fact what is now known as Ditylenchus dipsaci. It was first identified from diseased clover in England by Ritzema Bos in 1897 (Edwards, 1932) and was first recorded in lucerne in Britain in 1948 (Brown, 1953). Edwards (1932) states that Lounsbury reported it from South Africa in 1909 and also says that it may have been present for some years before this. McKay (1922) isolated Ditylenchus dipsaci from lucerne in Oregon and stated that it had not previously been recorded from this host in America. Godfrey (1922) reported that during the previous season it occurred in the United States of America in red clover, lucerne, strawberries and daffodils. Cobb (1923-29) recorded that Ditylenchus dipsaci was known to be widespread in the United States of America, occurring in twenty-six states all told. Noble (1925) called attention to the presence of the disease in lucerne in the Hunter River district of New South Wales, Australia, and stated that growers were of the opinion that Ditylenchus dipsaci had been present in their fields for many years. Edwards (1932) states that it was widespread throughout New South Wales.

Kirk (1903 and 1907) recorded this nematode in New Zealand from potatoes. Edwards (1932) stated that this record was not Ditylenchus dipsaci and Clark (1963) also questions it. Kirk (1908) published an
account of nematodes in New Zealand and in this the confusion over the potato nematode still persists, but *Ditylenchus dipsaci* was recorded from oats and wheat. Morrison (1957) was the first to record it in lucerne in New Zealand, but Smith and Palmer (pers. comm.) say that it was recognised before this. The distribution of this nematode in New Zealand is not well documented, but from the author's experience it is widespread throughout the Marlborough and Canterbury regions.

Ritzema Bos (Edwards, 1932) stated in 1888 that *Ditylenchus dipsaci* was known as a parasite of 34 species representing 14 natural orders of plants. Goodey (1929) lists it as occurring in 126 plant species representing 30 natural orders. In 1931 the same worker reported that the host range was 135 plants. Christie (1959) stated that the host range of *Ditylenchus dipsaci* was approaching 375 species. Altogether some 400 plants, both monocotyledons and dicotyledons, have been listed as hosts (Goodey, 1956 and Goodey et al., 1959).

Dingley (1969) made a thorough review of the host range in New Zealand and her list is as follows:

<table>
<thead>
<tr>
<th>Host</th>
<th>Plant Name</th>
</tr>
</thead>
<tbody>
<tr>
<td>Allium cepa</td>
<td>Onion</td>
</tr>
<tr>
<td>Avena sativa</td>
<td>Oats</td>
</tr>
<tr>
<td>Endymoin sp</td>
<td>English Bluebell</td>
</tr>
<tr>
<td>Gladiolus sp</td>
<td>Gladiolus</td>
</tr>
<tr>
<td>Hyacinthus orientalis</td>
<td>Hyacinth</td>
</tr>
<tr>
<td>Medicago sativa</td>
<td>Lucerne</td>
</tr>
<tr>
<td>Narcissus sp</td>
<td>Daffodils</td>
</tr>
<tr>
<td>Phlox paniculata</td>
<td>Phlox</td>
</tr>
<tr>
<td>Plantago lanceolata</td>
<td>Narrow leaved Plantain</td>
</tr>
<tr>
<td>Plantago major</td>
<td>Broad leaved Plantain</td>
</tr>
</tbody>
</table>

continued/...
3. BIOLOGICAL RACES

In 1888 Ritzema Bos grouped a number of nominal species together under the name of *Tylenchus devastatris* (the present *Ditylenchus dipsaci*) and at the same time noted that stem eelworms from different host plants showed different host preferences (Seinhorst, 1957). This was the first indication of the existence of biological races and since then many workers have investigated this phenomenon. Cross inoculation experiments by Goodey (1922), Hodson (1926) and others, have shown that these host preferences are constant and that the biological races are distinguishable biological units. Steiner (1925) in his discussion pointed out that different populations of a nematode species may vary in their relations to the same host species. He believed that a nematode population would always prefer that particular host species and the variety or strain in which its parents lived. He considered that this preference for one host species increased with the number of generations which had lived in it. When a population had grown long enough in a host plant he considered that it became so specialised that it could not easily attack other plant species.

Quanjer (1928) (Bingefors, 1957) thought that there were "bridging species" which made possible the transition from one plant species to

<table>
<thead>
<tr>
<th>Species</th>
<th>Common Name</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Secale cereale</em></td>
<td>Ryecorn</td>
</tr>
<tr>
<td><em>Solanum tuberosum</em></td>
<td>Potato</td>
</tr>
<tr>
<td><em>Trifolium pratense</em></td>
<td>Red Clover</td>
</tr>
<tr>
<td><em>Trifolium repens</em></td>
<td>White Clover</td>
</tr>
<tr>
<td><em>Triticum aestium</em></td>
<td>Wheat</td>
</tr>
<tr>
<td><em>Tulipa sp cult</em></td>
<td>Tulip</td>
</tr>
</tbody>
</table>
another. Goodey (1931) supposed a type of "food memory" as causing the preference of a nematode population for one host species.

Steiner (1956) (Eriksson, 1965) changed his ideas and said that he was of the opinion that most of the races are morphologically different and should be given specific rank. Eriksson (1965) noted that the Russian workers also held this view and had given specific names to some of the races. Seinhorst (1957) said that this idea of Steiners was not substantiated and he distinguished 11 races of *Ditylenchus dipsaci* each with its more or less restricted host range.

Sturhan (1964) using suitable host plants succeeded in interbreeding at least six biological races, in ten different combinations, substantiating that they were only races of the same nematode species. He obtained frequent occurrences of abnormalities in the shape of body and the tail in two hybrid populations, demonstrating the existence of remarkable genetical differences between the biological races. He suggested that the possibility of interbreeding is probably an important reason for the frequent occurrences of different races and of strains with different physiological behaviour within the biological races of one host plant species. It seems likely that new biological races could originate in this way.

Webster (1964) and Eriksson (1965) both managed to produce race hybrids using callous tissue cultures. Webster (1967) carried out crossing experiments with six different races of *Ditylenchus dipsaci* and produced 10 fertile hybrids. He tested the host range of five of these hybrids and found no relation between the host range of the hybrids and that of the parents, which contrasted with Eriksson's (1965) findings. He also found that on the average, the hybrids multiplied more slowly than their parents. Webster noted further that mixed populations must occur in nature, and his
results suggested that such mixtures would contain parental races, their hybrids and backcrosses. He says that back crossing, together with infertility or poor reproductive rate, may explain the slight variation in host range of the known races, and the absence of records of new races.

Webster (1967) sums up by saying, "At one time Ditylenchus dipsaci populations were probably separated geographically and ecologically on different natural hosts and so tend to form biological races that are host specific. Farming brought together these hosts and their nematode populations and created a dynamic situation with mixed populations of races, their hybrids and backcrosses, leading to populations with a host range spectrum differing slightly according to their genetical make up."

Smith (1951) recorded the presence of two biological races of Ditylenchus dipsaci that attacked lucerne.

In spite of all the investigations that have been carried out, it cannot be said that the problems of race differentiation and of the stability of the biological races of Ditylenchus dipsaci have been solved.

4. LIFE HISTORY

Very little work has been carried out on the life history of Ditylenchus dipsaci in its lucerne host. Yuskel (1960) however, gives an excellent description of its life history in onions at 15°C. and a brief resume of this follows.

The young Ditylenchus dipsaci females usually begin laying between three and seven days after the final moult, at a rate of eight to ten eggs per day, giving a range of 200 to 500 eggs per adult female. Webster (1967) substantiated this egg range. Immediately after laying, the eggs are
greyish in colour, but this colour pales with age to a very light grey hue. The first larval stage within the egg is thought to occur 5 - 5.5 days after egg deposition. The larvae is able to move slightly within the egg. Before this stage is reached no movement has been observed. This full grown first stage larvae moults almost immediately and the hatching of the second stage larvae from the egg takes place seven days after the beginning of the cycle.

The second moult occurs after another 2 - 2.5 days, the third moult after 3 - 3.5 days, and the fourth after another 4 - 5 days. Adult females and males were present 9 - 11 days from the time of egg hatch. One cycle in onions took between 19 and 23 days to complete at 15°C. and the longevity of both females and males ranged between 45 to 73 days.

The sex of the second stage larvae could not be determined in Yuskel's experiments but the sex of the third and fourth larvae stages could be ascertained by examination of the genital primordium. Females do not oviposit without being mated, nor do they continue to lay eggs without feeding (Palo 1962 and Webster 1967). A male may copulate with, and successfully fertilize, more than one female.

Fig. 1. shows an outline of the life history of *Ditylenchus dipsaci*.

The length of life of the stem nematode seems to depend very largely on environmental conditions. Hodson (1926) has shown that in the presence of moist, decaying tissue the organism dies within a very short period of time. It is possible, however, to keep adults and larvae alive for between four and five weeks at a temperature between 2°C. and 4°C.

Nematodes which leave the host tissue and migrate into the soil are unable to become quiescent while they remain in a moist environment and, moreover, they are unable to feed until they locate a suitable host. The
An outline of the Life History of *Ditylenchus dipsaci*. 
length of time during which this enforced starvation can be withstood is not known, but Goodey (1951) considers one year the maximum hostless period for *Ditylenchus dipsaci*.

However, in the absence of a moist environment, *Ditylenchus dipsaci* in common with many other nematodes, is able to become quiescent and remain in this dormant condition for long periods. Goodey (1923) showed that the pre-adult or fourth larval stage of *Ditylenchus dipsaci* was the form which appeared to resist desiccation for the longest periods. He found that quiescent nematodes from diseased onions and narcissus bulbs were readily revived in water after periods of desiccation ranging from 24 to 27 months. The same worker in 1930 showed that the period of desiccation for the larvae of *Ditylenchus dipsaci* was six years. Fielding (1951) (Van Grundy, 1965) sets the survival of *Ditylenchus dipsaci* in dried teasel at 20 - 23 years.

5. RESISTANCE TESTING TECHNIQUES

Determinations of resistance to *Ditylenchus dipsaci* may be carried out in different ways. The determination may be made by growing the plants to be investigated for resistance in field trials in naturally infected fields. Bingefors (1957) reviews this work and says that some workers compared the area infected with the total area covered by the variety, while others looked at the yield differences between infected and non-infected plots. (Bingefors in his work with red clover both charted the areas of infection and also measured yields.)

Efforts to produce more even infestations in field plots by spreading infected soil or plants over them were made by Frandsen (1951) on red clover
fields, and by Burkart (1937) (Bingefors, 1957) and Smith (1955) with lucerne.

The first trials in which plants were grown under relatively constant glasshouse conditions were those of Rostrup (1913) (Bingefors, 1957) in which soil from infested fields was used to provide the source of nematodes. Goodey (1922) used pots with infected soil and also added pieces of infected plants to ensure an overall uniform infection. He also used an inoculation technique in which nematode eggs were placed with a fine capillary glass pipette onto red clover plants that were at the first true leaf stage. Seinhorst (1945) (Bingefors, 1957) described a method whereby rye seedlings were inoculated with a number of nematodes in an incision in the coleoptile.

Bingefors (1957) states that his original laboratory technique for testing red clover for resistance to *Ditylenchus dipsaci* was to sow the red clover in boxes of soil infected with *Ditylenchus dipsaci*, or soil into which infected plants had been mixed. However, fungal attack on the plants was found to be difficult to overcome, so a modified method was used. The plants to be infected were grown in pots and infection was brought about by placing a folded piece of filter paper, which had been dipped in a suspension of nematodes in water, between the young shoots. After being inoculated the pots were placed in moist peatmould in a forcing pit covered by glass windows to produce a high humidity around the plant. This was found to be necessary during the first period of inoculation if even results were to be obtained. This technique was found to be very wasteful of space, to take a long time from inoculation to determination and not to give reliable results.
Bingefors (1957) in reviewing his earlier work discusses a seedling technique in which clover seeds were germinated and then as soon as possible transferred to filter paper strips. The seedlings were placed along the edge of one strip and covered with another. The two strips and plants were then rolled into a compact cylinder. Infection was originally carried out by dipping these rolls, seedlings downwards, into a suspension of nematodes containing 2000 - 3000 nematodes per ml. The plants remained immersed in the suspension for about an hour. The suspension was kept in motion by the passage of a weak air current. Frandsen (1951) modified this method and by means of a syringe dripped the nematode suspension onto the embryo plant before the filter papers were rolled together. The advantage of this method was the direct placement of the nematodes onto the plant and filter paper, thus providing favourable conditions for nematode movement and entry into the plant. Frandsen carried out tests with different numbers of nematodes in the inoculum and found that about 30 nematodes per drop gave the best results.

Bingefors (1957) states that in his earlier work he found that it was an advantage to apply the nematode suspension as a drop between the cotyledons with a pipette or hypodermic syringe. The best concentration here appeared to be about 2,500 - 3,500 nematodes per ml, or about 50 nematodes per drop, which was a suitable number to ensure infection. Dijkstra (1956) used this method of inoculation with only 15 - 20 nematodes per drop. He added carboxymethyl cellulose to the water to help prevent aggregation of the nematodes.

After inoculation, the plants were placed under cover to keep the humidity high and inoculation was repeated in all cases one or two days later. The inoculated rolls of seedlings were usually kept in pots or
beakers with water in a greenhouse with a temperature in the range of 12°C - 20°C. (Frandsen, 1951; Dijkstra, 1956; Bingefors, 1957).

The nematodes to be used in the inoculum by these workers were obtained by collecting infested plant material from the field and extracting the worms either from the fresh or dried material. Separation of the nematodes from the plant matter in these experiments (Frandsen, 1951; Dijkstra, 1956; Bingefors, 1957) was carried out with a modified Seinhorst apparatus, Seinhorst, (1950) (Bingefors, 1957) in which diseased plants are placed in large funnels and a fine mist of water is sprayed onto these plants. When the plants are wetted the nematodes leave the tissue and collect in receptacles at the base of the funnels. Frandsen scored his plants 10 - 15 days after the inoculation, on the swellings of the embryo stem. The plants were divided into responding (susceptible) and non-responding (resistant or presumably resistant). Any seedling that did not develop normally was disregarded.

Dijkstra (1956) cautions that in an assessment of resistant and susceptible plants all sorts of transitional stages can be found, making it difficult to distinguish susceptible and resistant plants. For further breeding work Dijkstra selected plants that were resistant (had no swelling) from his initial tests.

Bingefors (1957) carried out his initial assessment after two weeks and the plants were divided into resistant and susceptible categories according to their symptoms. Plants without swelling were considered resistant and plants with swelling were considered susceptible. He cautioned that some of the assessments did not give an entirely satisfactory result, but for direct selection he pointed out that one has to depend on visual symptoms because the plants must be retained alive. Bingefors (1957)
also stressed the importance of reproduction in the susceptible plants, that is the number of eggs that were produced by the nematodes in the plant. He recommended standardisation of the type of infected material that was used to provide the nematodes for inoculation and he used only dried material. Any plant that died was thought to be very susceptible because deaths were much higher in susceptible varieties than resistant ones.

In 10 week old plants Bingefors used the following scale to rate resistance.

- **0** = no nematodes
- **1** = very small number nematodes and no eggs
- **2** = a few nematodes and some eggs
- **3** = many nematodes and eggs or the plant was killed and possessed typical symptoms.

Bingefors then calculated an index of resistance from the formula

\[
\text{Index of resistance} = \left(1 - \frac{\sum a}{n}\right) \times 100
\]

where \( a \) is the classification of each individual plant

\( n \) is the number of observed plants

Most of this work was with red clover, but Bingefors did carry out some work with lucerne (Bingefors, 1961).

Frandsen (1951), Dijkstra (1956) and Bingefors (1957) all used artificial light in their greenhouses.

Grundbacher (1962) in his work with lucerne slightly modified the filter paper roll technique that had been used by the earlier workers. He germinated his seeds in moist vermiculite and then transferred them as seedlings to the filter paper rolls which were placed in growth cabinets at 11.1°C - 15.6°C, and at these temperatures he found that he could keep
them alive "for several months". The rolls were supplied with either tap water or a dilute Hoaglands solution and assessed for resistance after a month at 11.1°C or three weeks at 15.6°C. The strength of the inoculum used was 5,400 nematodes per ml and all the seedlings were inoculated twice. The resistant seedlings usually exhibited little swelling, but due to the presence of plants with intermediate swellings, Grundbacher maintains it was impossible to have an accurate classification on swellings alone. He found that the most critical evaluation of resistance was the amount of nematode reproduction taking place in the shoot apex region, even though reproduction could take place in the hypocotyl, the cotyledons and the petioles. Susceptible plants usually contained adult nematodes, eggs and some larvae in the shoot apex, while resistant plants had very few eggs present and the nematodes were smaller.

Grundbacher (1962) placed the seedlings in continuous light for an initial period after inoculation. He maintained that this prevented the cotyledons closing and facilitated the penetration of the nematodes into the plant. Hanna and Hawn (1965) who also used a filter paper roll technique, found that there was no necessity for a period of continuous light after inoculation.

Grundbacher (1962) stated that a visual assessment for resistance could be made by eliminating all plants that had any swelling. Stanford (pers. comm. 1969) believed that a swelling score for a series of plants within a variety would give a good evaluation of the resistance of that variety. He also said that in selection work, the swelling of the plants was a very satisfactory basis for division, provided an effective inoculation had been obtained. Because of this type of selection the level of resistance in the laboratory was found to be lower than the level of
resistance obtained in the field (Bingefors, 1957; Frandsen, 1951; Grundbacher, 1962).

Hanna and Hawn (1965) tested to determine the most suitable level of inoculum and found that adequate infestation was assured with 50 to 70 nematodes per drop. They finally used 40 to 50 nematodes per drop, applied on two successive days. They also found that using carboxymethyl cellulose as a carrier for nematodes had no significant effect upon the entry of nematodes into shoot apices. Griffin (1967) found that there was a greater percentage of plants galled when the cotyledons were inoculated with a methyl cellulose solution of nematodes, when the seedlings were maintained at 80% - 100% relative humidity, than when they were maintained at 30% - 60% relative humidity, and so established the necessity for high humidity that was used by earlier workers. He also found that an inoculum level of 50 nematodes per drop at the cotyledon stage was sufficient to give maximum galling.

Wynne and Busbice (1968), who grew the lucerne seedlings for inoculation in moist vermiculite, used a one to nine scale to rate the nematode damage to the plants, two weeks after inoculation. They then checked the infections by macerating the apical meristem and counting the nematodes and eggs present. They found that they could correlate swelling with susceptibility, but maintained that reproduction within the plant was a more direct criterion for susceptibility.

Temperature was found to influence the resistance of one lucerne variety. (Grundbacher and Stanford, 1962; Griffin, 1968) Lahontan became more susceptible as temperature rose, but the resistance of other varieties remained stable. Wynne and Busbice (1968) found that 19°C. appeared to be the most suitable temperature at which to carry out their tests.
It appears that nematodes enter all plants. In susceptible plants nematode multiplication is rapid, while in resistant plants there may be no reproduction at all (Bingefors, 1957; Grundbacher, 1962; Grundbacher and Stanford, 1962; Wynne and Busbice, 1968).
CHAPTER III

MATERIALS AND METHODS

1. NEMATODE COLLECTION, PRESERVATION AND EXTRACTION

The symptoms of *Ditylenchus dipsaci* attack on lucerne are most recognisable in Canterbury during the spring (September and October) and autumn (March and April). The typical field symptoms at these times are circular or oval patches where plants have been killed or badly stunted. These affected areas make very little growth, are usually very slow in recovering after cutting, and invariably fail to show any appreciable new growth in the following spring. The affected plants present a typical dwarfed and distorted appearance. There is a marked reduction in the number of stems and these are swollen and generally show a distinct brownish colouration especially at the base, although in many cases it extends up the stem for some distance. Generally, affected stems are very brittle and easily broken off. The new buds arising from the crowns of affected plants are usually swollen, spongy in texture and pale greenish brown in colour. In cases of severe infestation, the crowns rot away and the plants eventually die out, leaving a stand of unprofitable thinness which rapidly becomes overgrown with weeds and grasses.

Infected stands of lucerne on properties at North Loburn and Rakaia were used as the sources of nematode infected plants. The stunted plants were harvested either by hand or with a small rotary mower. This material was then placed in cotton bags and carried back to Lincoln College where it was dried in ovens set at 30°C. for 24 hours. This temperature killed all
nematodes, apart from the fourth larval stage. It was found that this was more satisfactory than air drying the material, due to a tendency of the material to heat and rot. This material, once dry, was stored in cotton bags in a dry place until inoculum was required.

The apparatus used to separate the nematodes from the dried lucerne is illustrated in Figure 2. It consisted of a two foot cube metal waterproof container. This container was mounted on legs to produce a fall of one and a half inches from the rear to the forward edge, and three outlets were incorporated at six, twelve and eighteen inches along its lower forward edge. Brackets were provided in the container to hold a nylon gauze covered wooden frame. A spray nozzle TN3V manufacturer's number (Spraying Systems Co.), with a spray rate of 3.75 gallons per hour at a water pressure of 60 lbs. per square inch, was positioned on an adjustable bracket above the container. The spray nozzle was at the end of a water supply line in which the water flow was regulated by a solenoid controlled valve activated by a time switch which opened the solenoid control valve for one minute in every four minutes. This diminished the nozzle's rated flow to under one gallon per hour. Three plastic funnels of six inch diameter with clamp sealed ends were positioned below the outlets, so that the run off water could be retained. As water was allowed to overflow these funnels the whole apparatus had to be positioned on a freely draining floor.

The infected dried lucerne was spread on the gauze covered frame. The action of dampening the infected material revives the quiescent fourth larval stage nematodes, which move with the water through the outlets of the container into the funnels in which they settle under the influence of gravity. The low rate of water flow used was to prevent the possibility
Apparatus for the extraction of nematodes from plant material.
of the nematodes being carried away with the overflowing water from the funnels.

After 24 hours, nematodes could be drawn off from the funnels. It was found that the best draw off of nematodes was after 36 hours, and that even after seven days it was still possible to obtain nematodes from the one sample of lucerne. Nematodes extracted after 36 hours were usually sluggish due to the lack of oxygen in the water. These were revived by treating with an aerator, which simply bubbled air through the suspension of nematodes, giving them additional oxygen.

Soil and plant material were also carried into the funnels and were separated from the nematodes by filtering through a coarse mesh sieve covered with toilet paper, as shown in Figure 3. The suspension containing the nematodes and extraneous material was poured into the sieve, which was positioned in a dish in such a manner that the water level reached the toilet paper. This enabled the nematodes to swim through, leaving the debris behind on the upper surface of the paper.

A second method was to pour the suspension of nematodes, soil and plant material onto a filter paper in a filter funnel. The nematodes and the debris were retained on the filter paper. The paper was then laid over an inverted watch glass within a petri dish, and water was added until it just lapped the filter paper. The nematodes would then move down the filter paper to the water, while the extraneous matter remained held on the filter paper (Fig. 4). Neither method was found to be superior and both gave satisfactory results. The nematodes were then allowed to sink to the bottom of the vessel and excess water was decanted.

1 Jeyes Baby Soft Toilet Paper.
Apparatus for separating nematodes from soil and plant debris.
Filter paper with nematodes and rubbish

Watch glass

Petri dish

Water level

Apparatus for separating nematodes from soil and plant debris
The nematodes were stored in water at 2°C - 4°C until needed. The storage time was normally for one or two days only, but viable nematodes have been kept as long as four weeks at this temperature.

2. **PLANT PREPARATION**

The lucerne seedlings used during this project were grown in rolls of filter paper or in soil flats.

The seeds of all varieties (lines) used during this work were surface sterilized before germination. The seed was soaked in 30% hydrogen peroxide for one hour and then, without rinsing, were placed on filter paper in petri dishes to germinate. The advantage of this method was that there was no necessity to rinse the seed with water, and so there was very little chance of contamination after the sterilization. It was found that this method did not give a complete kill of seed coat carried pathogens, and consequently a second method, which consisted of treating the seed with a solution composed of equal parts of 1N HCL and commercial bleach\(^1\) for 12 minutes, was used. This was found to be satisfactory, even though rinsing with distilled water had to be carried out, thus increasing the possibility of recontamination.

A test germination to estimate the proportion of diseased seed was undertaken. Due to the difficulties experienced in controlling disease of the seedlings, any seed line with a high level of contamination was rejected.

The germination of the seed for use in the filter paper rolls was carried out in covered petri dishes on a double layer of filter paper

\(^1\) Commercial bleach used was Janola.
soaked with water containing the fungicide captan.

Many more seeds than necessary were germinated so that selection could be made for seedlings that were at the same stage of development. This selection was made after two to three days and the seedlings chosen were transferred to filter paper rolls.

Filter paper rolls were made from strips 11.5 cm. x 28.5 cm. of Whatman's chromatography paper No. 1 (soft) and No. 50 (hard). The lucerne seedlings were placed on a strip of hard paper, moistened with a captan solution, so that their cotyledons extended above the margins of the paper. The strips were rolled using a glass tube for guidance, and the roll was secured with a rubber band. Fifteen seedlings were grown per roll. A method employing 50 seedlings on strips 11.5 cm. x 57 cm. was found to be unsatisfactory due to the large number of seedlings requiring attention at inoculation.

The completed rolls were then placed in plastic pots of 1.5 in. in diameter and 2 in. in height. Water was added to these pots to provide moisture for the first seven days, and then a Hoaglands solution (Hoagland and Arnon, 1950) was added to provide nutrients for continued growth.

The completed pots were placed in trays in a growth cabinet which provided a 16 hour day at a temperature of 16°C. The trays were covered by plastic sheets which were supported by a frame manufactured from eight gruge wire (Plate 1.). These frames had to be high enough to allow ample room for the handling of the pots. The trays contained water so that the seedlings in the rolls were held at a relative humidity approaching 100. This was to facilitate the entry of the nematodes into the plant after inoculation.
PLATE 1

Covered trays used to hold the filter paper rolls in the growth cabinet
The seedlings were then left until the development of the first true leaf stage. The inoculations were then carried out. This was normally about two days after the seedlings had been placed in the rolls.

The seedlings and the rolls were sprayed with a small amount of a captan solution every day throughout the experiments, to help prevent fungal attack.

The seedlings grown in soil were raised in flats 15 in. x 18 in., each sown with 100 lucerne seeds which had been surface sterilized by the same method as previously described. The seed had then been air dried and rubbed with captan as an added precaution against fungal attack.

The seeds were planted in 10 rows of 10 each. The spacing was obtained by using a board the same area as the surface of the flat, with 100 lead head nails driven into it. This could be pressed into the soil in the flat and the 100 indentations obtained were evenly spaced.

In each of these 10 rows, five of the indentations were sown with single seeds, and the remaining five were sown with three seeds each, to allow for seedling mortality. Transplanting took place where necessary as soon as the seedlings had fully developed cotyledons. Extra seedlings that were not required for transplanting were retained, because at this stage there would be no competition between plants.

The flats were placed in trays in a glasshouse, the temperature of which varied daily between 16°C - 29°C. Artificial lighting in this glasshouse was controlled by a time switch to operate between seven a.m. and seven p.m. Overhead watering of the seedlings was avoided by standing the flats in metal trays. This allowed the seedlings to draw water as required.
The seedlings were inoculated at the first true leaf stage and again two days later. The relative humidity about the plants at inoculation was increased towards the 100% level by covering the flats and the trays with transparent plastic sheeting supported by frames of eight gauge wire.

The seedlings in the flats were sprayed with a small amount of captan daily to reduce fungal attack.

3. PLANT INOCULATION

Clean nematodes that had been stored under refrigeration (pp.25) were revived by aerating the water in which they were stored. Methyl cellulose was added to increase the viscosity of the solution. This helped to retain the nematodes on the seedlings, thus enhancing their infective ability. It also tended to slow the aggregation of the nematodes, and so the number of nematodes in the inoculum was more constant from the beginning to the end of an inoculation series.

Both three and one percent solutions of methyl cellulose were used; however, it was found that the one percent solution was the better, due to the tendency of the three percent solution to form a hard skin of methyl cellulose which inhibited the growth of the seedling.

The inoculum was applied to the seedling by the use of a syringe with an 18 gauge needle (Plate 2). One drop of inoculum was added to each plant, the drop being positioned between the cotyledons at the apical region. The number of nematodes in a sample of ten drops of inoculum was determined by examination under a stereoscopic microscope, and the number of nematodes in the solution was adjusted until each drop contained approximately 50 nematodes.
PLATE 2

Syringe used for inoculation of lucerne seedlings.
The inoculation of the seedlings on the rolls was carried out in a sterile cabinet. The rolls were then replaced in the humidity chamber in the growth cabinet, left for two days, and then reinoculated. Each seedling received, therefore, a total inoculation of approximately 100 nematodes over a time period of two days.

The seedlings in soil flats were inoculated in the glasshouse. The flats were covered with plastic sheets as described, and then left for two days before reinoculation.

It was found that during inoculations it was necessary to keep the inoculum slightly chilled and also to stir it frequently to stop any aggregation of the nematodes. The aerator could not be used for stopping aggregation because it caused too many bubbles in the inoculum solution.

After inoculation the plants were left for three weeks (Plate 3) and their symptoms (swelling in the apical region) were assessed either visually, or by measuring the width of the stem between the cotyledons (Plate 4). This measurement was carried out with an eyepiece graticule in a stereoscopic microscope. A sample of seedlings was then macerated individually and a check was made for the presence of nematodes and eggs.

All the preliminary work in this research project was carried out using the lucerne variety Wairau. This is the most common variety of lucerne grown in New Zealand, but no information is available on its resistance to stem nematode.

In the experiments where the relative resistance of lucerne varieties to stem nematodes was determined, three lucerne varieties were used. These were Wairau, Grimm and Washoe. Grimm is an old variety of lucerne which is known to be highly susceptible to attack by stem nematode (Bingefors, 1961). Washoe, which was released by the United States Department of Agriculture in
PLATE 3

Lucerne plants in filter paper rolls three weeks after inoculation

Key:  
- c - control  
- i - infected
PLATE 4

Nematode infected and uninfected lucerne plants after three weeks

Top line - Infected

- Swollen stem

Bottom line - Uninfected
1966 (Hunt, et al, 1966), is an eight clone synthetic variety derived mainly of Nemstan and known to have a high degree of resistance to stem nematode.
CHAPTER IV

RESULTS

1. RESULTS FROM SOIL FLAT TRIALS

Although eventually it was possible to successfully grow lucerne seedlings in filter paper rolls, initially the extensive losses due to fungal attack caused some doubt as to whether this method could be used in this study at all. It was decided, therefore, to set up a trial in soil filled flats to find out something about the efficiency of inoculation techniques, the effect, if any, of using a methyl cellulose solution on the seedlings, and the accuracy of judging whether or not infection of the seedlings had taken place.

Twelve flats were sown with Wairau lucerne by the method described, and grouped as follows. (A replication consisted of 200 plants).

(i) Flats 1 and 2 (Control) were not inoculated.

(ii) Flats 3 and 4 (Methyl cellulose) were inoculated with one percent solution of methyl cellulose alone.

(iii) Flats 5 to 12 (Replications 1 to 4) were inoculated with nematodes in a one percent solution of methyl cellulose.

Three estimates of infection of the seedlings by the nematodes were made; the first after two weeks, the second at three weeks, and the third at four weeks. These estimates were carried out on the condition of the apical region of the plant; plants with swollen stems being regarded as infected. The results of the test are tabulated in Tables 1, 2 and 3.
About 65 percent of the seedlings became infected, 35 percent remained uninfected. Losses from causes other than nematodes approximated those of seedlings in control flats.

The plants were grown for a further month in the flats after these tests were finished, and the ability of some of the stunted plants to recover was noted. At times a lateral shoot became the main growing stem, leaving the apical stem stunted. This meant that although the plants harboured a population of nematodes, they did not have overall stunting.

TABLE 1

| Condition of Young Lucerne Plants (Wairau) Two Weeks After Infection with Nematodes in 1% Solution of Methyl Cellulose |
|---|---|---|---|
| | Infected | Normal | Died | Total |
| Control | 0 | 255 | 2 | 257 |
| Methyl Cellulose | 0 | 255 | 5 | 260 |
| Rep. 1 | 109 | 184 | 4 | 297 |
| Rep. 2 | 142 | 125 | 5 | 272 |
| Rep. 3 | 122 | 165 | 0 | 289 |
| Rep. 4 | 134 | 127 | 4 | 265 |
### TABLE 2

**CONDITION OF YOUNG LUCERNE PLANTS (WAIRAU) THREE WEEKS AFTER INFECTION WITH NEMATODES IN 1% SOLUTION OF METHYL CELLULOSE.**

<table>
<thead>
<tr>
<th>Plant Numbers</th>
<th>Infected</th>
<th>Normal</th>
<th>Died</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0</td>
<td>253</td>
<td>4</td>
<td>257</td>
</tr>
<tr>
<td>Methyl Cellulose</td>
<td>0</td>
<td>252</td>
<td>8</td>
<td>260</td>
</tr>
<tr>
<td>Rep. 1</td>
<td>175</td>
<td>113</td>
<td>9</td>
<td>297</td>
</tr>
<tr>
<td>Rep. 2</td>
<td>144</td>
<td>112</td>
<td>6</td>
<td>262</td>
</tr>
<tr>
<td>Rep. 3</td>
<td>157</td>
<td>131</td>
<td>1</td>
<td>289</td>
</tr>
<tr>
<td>Rep. 4</td>
<td>154</td>
<td>105</td>
<td>6</td>
<td>265</td>
</tr>
</tbody>
</table>

### TABLE 3

**CONDITION OF YOUNG LUCERNE PLANTS (WAIRAU) FOUR WEEKS AFTER INFECTION WITH NEMATODES IN 1% SOLUTION OF METHYL CELLULOSE.**

<table>
<thead>
<tr>
<th>Plant Numbers</th>
<th>Infected</th>
<th>Normal</th>
<th>Died</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0</td>
<td>249</td>
<td>9</td>
<td>260</td>
</tr>
<tr>
<td>Methyl Cellulose</td>
<td>0</td>
<td>251</td>
<td>8</td>
<td>257</td>
</tr>
<tr>
<td>Rep. 1</td>
<td>183</td>
<td>103</td>
<td>11</td>
<td>297</td>
</tr>
<tr>
<td>Rep. 2</td>
<td>153</td>
<td>110</td>
<td>9</td>
<td>272</td>
</tr>
<tr>
<td>Rep. 3</td>
<td>209</td>
<td>78</td>
<td>2</td>
<td>289</td>
</tr>
<tr>
<td>Rep. 4</td>
<td>167</td>
<td>92</td>
<td>6</td>
<td>265</td>
</tr>
</tbody>
</table>
2. RESULTS FROM FILTER PAPER ROLL TRIALS

In the filter paper roll trials a series of experiments, Nos. 1 - 13, were carried out and these are dealt with in sequence.

**Experiment 1**

The first experiment using filter paper rolls to hold the seedlings employed strips of Whatmans No. 1 and No. 50 chromatography paper, 11.5 cm. x 57 cm. Each roll contained 50 seedlings. Nine rolls were made, one of these being kept as a control. The seedlings in the remaining eight rolls were inoculated twice, each inoculation drop containing approximately 50 nematodes suspended in a 3% solution of methyl cellulose. These rolls were then left for three weeks in the growth cabinet at 16°C. and then assessed on the basis that swollen plants were susceptible and non swollen plants were normal. To check these results the plants were macerated in water in a watch glass and examined under a stereoscopic microscope for nematodes and eggs. The results are given in Table 4.
TABLE 4

RESULTS FROM FILTER PAPER ROLLS IN EXPERIMENT 1

<table>
<thead>
<tr>
<th>Roll No.</th>
<th>Initial No. of Plants</th>
<th>Survival after 3 weeks</th>
<th>No. Plants killed by fungal attack</th>
<th>No. Plants showing nematode symptoms</th>
<th>No. Plants containing nematodes</th>
<th>Normal Plants</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (Control)</td>
<td>50</td>
<td>39</td>
<td>11</td>
<td>0</td>
<td>0</td>
<td>39</td>
</tr>
<tr>
<td>2</td>
<td>50</td>
<td>16</td>
<td>34</td>
<td>14</td>
<td>14</td>
<td>2</td>
</tr>
<tr>
<td>3</td>
<td>50</td>
<td>16</td>
<td>34</td>
<td>16</td>
<td>16</td>
<td>--</td>
</tr>
<tr>
<td>4</td>
<td>50</td>
<td>5</td>
<td>45</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>5</td>
<td>50</td>
<td>34</td>
<td>16</td>
<td>31</td>
<td>30</td>
<td>3</td>
</tr>
<tr>
<td>6</td>
<td>50</td>
<td>15</td>
<td>35</td>
<td>15</td>
<td>15</td>
<td>--</td>
</tr>
<tr>
<td>7</td>
<td>50</td>
<td>8</td>
<td>42</td>
<td>8</td>
<td>8</td>
<td>--</td>
</tr>
<tr>
<td>8</td>
<td>50</td>
<td>10</td>
<td>40</td>
<td>10</td>
<td>10</td>
<td>--</td>
</tr>
<tr>
<td>9</td>
<td>50</td>
<td>31</td>
<td>19</td>
<td>26</td>
<td>26</td>
<td>5</td>
</tr>
</tbody>
</table>

Of the total of 400 nematode treated plants, 135 survived and 120 of these plants had symptoms of nematode attack. (Roll 4 was eliminated).

Of these 120, when macerated in water, 119 had nematodes and eggs present. One plant had nematodes but no eggs. The 10 plants that had no swelling had no nematodes present when macerated.

Experiments 2 - 8

Seven experiments (Nos. 2 - 8) were attempted but all were unsuccessful because of seedling mortality. Fungal attack caused the majority of the deaths. Surface sterilization of the seed was carried out (pp.25). The temperature in the growth cabinet was lowered to 12°C to try and help prevent
the fungal attack, but without success. The three main pathogens that attacked the seedlings were *Aspergillus niger*, *Phoma medicaginis* and mildew (*Peronospora trifoliorum*). The seed line of Wairau used in these trials was tested by the Plant Disease Division, D.S.I.R., Lincoln, and found to carry a very heavy infection of the seed born pathogen *Phoma medicaginis*. This line was discarded and a clean seed line was obtained for the rest of the research work.

In experiments 2 - 8 it was realised that 50 plants per roll were too many, because the paper size used in the rolls (11.5 cm. x 57 cm.) was too large to work with efficiently. In addition to this the 3% solution of methyl cellulose caused the plants to stick together, and also caused the binding of the apical region, thus inhibiting growth. The number of seedlings was subsequently reduced to 15 per roll and a 1% solution of methyl cellulose was used instead of the 3% solution.

**Experiment 9**

Forty filter paper rolls were made from papers 11.5 cm. x 22.5 cm. with 15 seedlings of Wairau lucerne in each. These were grouped as follows:

(i) Rolls 1 - 14 were inoculated with nematodes in a 1% solution of methyl cellulose.

(ii) Rolls 15 - 27 were inoculated with 1% methyl cellulose alone.

(iii) Rolls 28 - 40 were not inoculated.

Rolls 1 - 27 were inoculated by the method described. All the seedlings were supplied with Hoaglands nutrient solution after seven days of being in the rolls, and sprayed with a solution of captan daily. The seedlings were then left for three weeks in a growth cabinet at 16°C and assessment of infection was carried out visually. The results are given in Tables 5, 6 and 7.
<table>
<thead>
<tr>
<th>Roll No.</th>
<th>Initial Plant Numbers</th>
<th>Number Plants Surviving</th>
<th>Mortality</th>
<th>No. Plants showing nematode symptoms</th>
<th>Normal Plants</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>15</td>
<td>14</td>
<td>1</td>
<td>14</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>15</td>
<td>12</td>
<td>3</td>
<td>12</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>15</td>
<td>15</td>
<td>0</td>
<td>15</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>15</td>
<td>14</td>
<td>1</td>
<td>14</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>15</td>
<td>12</td>
<td>3</td>
<td>12</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>15</td>
<td>14</td>
<td>1</td>
<td>14</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>15</td>
<td>15</td>
<td>0</td>
<td>15</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>15</td>
<td>15</td>
<td>0</td>
<td>15</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>15</td>
<td>13</td>
<td>2</td>
<td>13</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>15</td>
<td>14</td>
<td>1</td>
<td>14</td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>15</td>
<td>14</td>
<td>1</td>
<td>14</td>
<td></td>
</tr>
<tr>
<td>12</td>
<td>15</td>
<td>15</td>
<td>0</td>
<td>15</td>
<td></td>
</tr>
<tr>
<td>13</td>
<td>15</td>
<td>12</td>
<td>3</td>
<td>11</td>
<td>1</td>
</tr>
<tr>
<td>14</td>
<td>15</td>
<td>15</td>
<td>0</td>
<td>15</td>
<td></td>
</tr>
<tr>
<td>Totals:</td>
<td>210</td>
<td>194</td>
<td>16</td>
<td>193</td>
<td>1</td>
</tr>
</tbody>
</table>
There was no obvious swelling in any seedlings in the remaining rolls so mortalities only were recorded:

**TABLE 6**

<table>
<thead>
<tr>
<th>Roll No.</th>
<th>Initial Plant Nos.</th>
<th>Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>15</td>
<td>0</td>
</tr>
<tr>
<td>16</td>
<td>15</td>
<td>1</td>
</tr>
<tr>
<td>17</td>
<td>15</td>
<td>3</td>
</tr>
<tr>
<td>18</td>
<td>15</td>
<td>4</td>
</tr>
<tr>
<td>19</td>
<td>15</td>
<td>2</td>
</tr>
<tr>
<td>20</td>
<td>15</td>
<td>1</td>
</tr>
<tr>
<td>21</td>
<td>15</td>
<td>0</td>
</tr>
<tr>
<td>22</td>
<td>15</td>
<td>3</td>
</tr>
<tr>
<td>23</td>
<td>15</td>
<td>1</td>
</tr>
<tr>
<td>24</td>
<td>15</td>
<td>1</td>
</tr>
<tr>
<td>25</td>
<td>15</td>
<td>2</td>
</tr>
<tr>
<td>26</td>
<td>15</td>
<td>2</td>
</tr>
<tr>
<td>27</td>
<td>15</td>
<td>0</td>
</tr>
</tbody>
</table>

**Totals:** 195 21
TABLE 7

MORTALITIES IN THE CONTROL ROLLS

<table>
<thead>
<tr>
<th>Roll No.</th>
<th>Initial Plant Nos.</th>
<th>Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>28</td>
<td>15</td>
<td>0</td>
</tr>
<tr>
<td>29</td>
<td>15</td>
<td>1</td>
</tr>
<tr>
<td>30</td>
<td>15</td>
<td>3</td>
</tr>
<tr>
<td>31</td>
<td>15</td>
<td>4</td>
</tr>
<tr>
<td>32</td>
<td>15</td>
<td>2</td>
</tr>
<tr>
<td>33</td>
<td>15</td>
<td>1</td>
</tr>
<tr>
<td>34</td>
<td>15</td>
<td>0</td>
</tr>
<tr>
<td>35</td>
<td>15</td>
<td>3</td>
</tr>
<tr>
<td>36</td>
<td>15</td>
<td>1</td>
</tr>
<tr>
<td>37</td>
<td>15</td>
<td>1</td>
</tr>
<tr>
<td>38</td>
<td>15</td>
<td>2</td>
</tr>
<tr>
<td>39</td>
<td>15</td>
<td>2</td>
</tr>
<tr>
<td>40</td>
<td>15</td>
<td>0</td>
</tr>
<tr>
<td>Totals:</td>
<td>195</td>
<td>21</td>
</tr>
</tbody>
</table>

Experiment 10

Fifteen filter paper rolls with 15 lucerne seedlings in each were set up as follows:

(i) Rolls 1 - 5 contained seedlings of the lucerne variety Wairau. The lucerne seedlings in Roll 1 were not inoculated. The lucerne seedlings in rolls 2 - 5 were inoculated with nematodes in a 1% solution of methyl cellulose.
(ii) Rolls 6 - 10 contained seedlings of the lucerne variety Washoe. The lucerne seedlings in Roll 6 were not inoculated. The lucerne seedlings in Rolls 7 - 10 were inoculated with nematodes in a 1% solution of methyl cellulose.

(iii) Rolls 11 - 15 contained seedlings of the lucerne variety Lahontan. The lucerne seedlings in Roll 11 were not inoculated. The lucerne seedlings in Rolls 12 - 15 were inoculated with nematodes in a 1% solution of methyl cellulose.

All seedlings were grown for a period of three weeks in a growth cabinet, and again assessment of infection was visual. The results are given in Table 8.

**TABLE 8**

RESULTS FROM THREE LUCERNE VARIETIES TESTED FOR NEMATODE SUSCEPTIBILITY

<table>
<thead>
<tr>
<th>Roll No.</th>
<th>Initial Plant Numbers</th>
<th>Number Plants Surviving</th>
<th>Mortality</th>
<th>No. Plants showing nematode attack</th>
<th>Normal Plants</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (Control)</td>
<td>15</td>
<td>10</td>
<td>5</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>15</td>
<td>12</td>
<td>3</td>
<td>11</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>15</td>
<td>13</td>
<td>2</td>
<td>13</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>15</td>
<td>15</td>
<td>0</td>
<td>15</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>15</td>
<td>11</td>
<td>4</td>
<td>11</td>
<td></td>
</tr>
<tr>
<td>Totals:</td>
<td>75</td>
<td>61</td>
<td>14</td>
<td>50</td>
<td></td>
</tr>
</tbody>
</table>

continued/...
TABLE 8 (Continued)

\textbf{Washoe} \begin{tabular}{lcccc}
\hline
\textbf{Roll No.} & \textbf{Initial Plant Numbers} & \textbf{Number Plants Surviving} & \textbf{Mortality} & \textbf{No. Plants showing nematode attack} & \textbf{Normal Plants} \\
\hline
6 (Control) & 15 & 12 & 3 & 0 & \\
7 & 15 & 13 & 2 & 13 & \\
8 & 15 & 15 & 0 & 15 & \\
9 & 15 & 8 & 7 & 8 & \\
10 & 15 & 9 & 6 & 7 & 2 \\
\hline
\textbf{Totals:} & 75 & 57 & 18 & 43 & 2 \\
\end{tabular}

\textbf{Lahontan} \begin{tabular}{lcccc}
\hline
\textbf{Roll No.} & \textbf{Initial Plant Numbers} & \textbf{Number Plants Surviving} & \textbf{Mortality} & \textbf{No. Plants showing nematode attack} & \textbf{Normal Plants} \\
\hline
11 (Control) & 15 & 10 & 5 & 0 & \\
12 & 15 & 11 & 4 & 11 & \\
13 & 15 & 14 & 1 & 13 & 1 \\
14 & 15 & 9 & 6 & 9 & \\
15 & 15 & 7 & 8 & 7 & \\
\hline
\textbf{Totals:} & 75 & 51 & 24 & 40 & 1 \\
\end{tabular}
Experiment 11

Fifty-four filter paper rolls with 15 lucerne seedlings in each were set up as follows: (A unit was taken as 45 plants and thus made up of three rolls.)

(i) Rolls 1 - 18 contained seedlings of the lucerne variety Washoe. The seedlings in Rolls 1 - 6 (controls) were not inoculated. The seedlings in Rolls 7 - 18 (treatments) were inoculated with nematodes in a 1% solution of methyl cellulose.

(ii) Rolls 19 - 36 contained seedlings of the lucerne variety Wairau. The seedlings in Rolls 19 - 24 (controls) were not inoculated. The seedlings in Rolls 25 - 36 (treatments) were inoculated with nematodes in a 1% solution of methyl cellulose.

(iii) Rolls 37 - 54 contained seedlings of the lucerne variety Grimm. The seedlings in Rolls 37 - 42 (controls) were not inoculated. The seedlings in Rolls 43 - 54 (treatments) were inoculated with nematodes in a 1% solution of methyl cellulose.

All seedlings were then left for three weeks in a growth cabinet at 16°C. The results of this experiment are given in Table 9. Assessment was by measurement of the stem diameter and the measurements for the whole sample are given in Appendix 1.
TABLE 9

RESULTS FROM THREE LUCERNE VARIETIES
TESTED FOR NEMATODE SUSCEPTIBILITY

<table>
<thead>
<tr>
<th></th>
<th>Initial No. of Plants</th>
<th>Plants Surviving</th>
<th>Mortality</th>
<th>Mean Stem Diameter in mm.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Washoe</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Controls</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 1-3</td>
<td>45</td>
<td>40</td>
<td>5</td>
<td>2.72</td>
</tr>
<tr>
<td>Rolls 4-6</td>
<td>45</td>
<td>35</td>
<td>10</td>
<td>2.83</td>
</tr>
<tr>
<td>Treatments</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 7-9</td>
<td>45</td>
<td>35</td>
<td>10</td>
<td>3.98</td>
</tr>
<tr>
<td>Rolls 10-12</td>
<td>45</td>
<td>37</td>
<td>8</td>
<td>3.76</td>
</tr>
<tr>
<td>Rolls 13-15</td>
<td>45</td>
<td>36</td>
<td>9</td>
<td>3.88</td>
</tr>
<tr>
<td>Rolls 16-18</td>
<td>45</td>
<td>29</td>
<td>16</td>
<td>3.79</td>
</tr>
<tr>
<td>Totals:</td>
<td>270</td>
<td>212</td>
<td>58</td>
<td></td>
</tr>
</tbody>
</table>

Whirau       

<table>
<thead>
<tr>
<th></th>
<th>Initial No. of Plants</th>
<th>Plants Surviving</th>
<th>Mortality</th>
<th>Mean Stem Diameter in mm.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Controls</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 19-21</td>
<td>45</td>
<td>45</td>
<td>0</td>
<td>2.96</td>
</tr>
<tr>
<td>Rolls 22-24</td>
<td>45</td>
<td>44</td>
<td>1</td>
<td>3.02</td>
</tr>
<tr>
<td>Treatments</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 25-27</td>
<td>45</td>
<td>42</td>
<td>3</td>
<td>4.53</td>
</tr>
<tr>
<td>Rolls 28-30</td>
<td>45</td>
<td>42</td>
<td>3</td>
<td>4.58</td>
</tr>
<tr>
<td>Rolls 31-33</td>
<td>45</td>
<td>41</td>
<td>4</td>
<td>4.57</td>
</tr>
<tr>
<td>Rolls 34-36</td>
<td>45</td>
<td>38</td>
<td>7</td>
<td>4.77</td>
</tr>
<tr>
<td>Totals:</td>
<td>270</td>
<td>252</td>
<td>18</td>
<td></td>
</tr>
</tbody>
</table>
TABLE 9 (Continued)

<table>
<thead>
<tr>
<th></th>
<th>Initial No. of Plants</th>
<th>Plants Surviving</th>
<th>Mortality</th>
<th>Mean Stem Diameter in mm.</th>
</tr>
</thead>
</table>

**Grimm**

**Controls**

<table>
<thead>
<tr>
<th>Rolls 37-39</th>
<th>45</th>
<th>37</th>
<th>8</th>
<th>2.98</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rolls 40-42</td>
<td>45</td>
<td>40</td>
<td>5</td>
<td>3.03</td>
</tr>
</tbody>
</table>

**Treatments**

<table>
<thead>
<tr>
<th>Rolls 43-45</th>
<th>30¹</th>
<th>27</th>
<th>3</th>
<th>5.21</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rolls 46-48</td>
<td>45</td>
<td>41</td>
<td>4</td>
<td>5.40</td>
</tr>
<tr>
<td>Rolls 49-51</td>
<td>45</td>
<td>35</td>
<td>10</td>
<td>4.97</td>
</tr>
<tr>
<td>Rolls 52-54</td>
<td>45</td>
<td>37</td>
<td>8</td>
<td>5.38</td>
</tr>
</tbody>
</table>

**Totals:**

255                   217               38

¹ Roll 44 dried out and the plants could not be measured.

A sample of the measured plants from each of the three varieties was macerated with water and examined to see whether there were nematodes (larvae and adults bulked) and eggs present. The means of this sample are given in Table 10. (The plants used are categorized in Appendix 2).
### TABLE 10

RESULTS FROM THE MACERATED LUCERNE SEEDLINGS FROM EXPERIMENT 11

<table>
<thead>
<tr>
<th>Variety</th>
<th>Washoe</th>
<th>Wairau</th>
<th>Grimm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of plants tested</td>
<td>73</td>
<td>131</td>
<td>87</td>
</tr>
<tr>
<td>Number of plants with nematodes and eggs</td>
<td>7</td>
<td>70</td>
<td>46</td>
</tr>
<tr>
<td>Average stem diameter of plants with nematodes and eggs (in mm.)</td>
<td>4.96</td>
<td>5.43</td>
<td>6.43</td>
</tr>
<tr>
<td>Number of plants with nematodes only</td>
<td>51</td>
<td>32</td>
<td>12</td>
</tr>
<tr>
<td>Average stem diameter of plants with nematodes only (in mm.)</td>
<td>3.92</td>
<td>4.03</td>
<td>5.00</td>
</tr>
<tr>
<td>Plants with no attack at all</td>
<td>15</td>
<td>29</td>
<td>24</td>
</tr>
<tr>
<td>Average stem diameter of plants with no attack (in mm.)</td>
<td>2.84</td>
<td>3.22</td>
<td>3.36</td>
</tr>
</tbody>
</table>

If the ability of the nematodes to reproduce within a plant is a sign that the plant is susceptible to attack, then the percentage of plants containing reproducing nematodes should give an indication of the susceptibility of the variety.

These percentages are as follows:

- Washoe 9.6%
- Wairau 53.4%
- Grimm 52.9%
Experiment 12

Thirty-six filter paper rolls with 15 lucerne seedlings in each were set up as follows: (A unit was taken as 45 plants and thus made up of three rolls.)

(i) Rolls 1 - 12 contained seedlings of the lucerne variety Washoe. The seedlings in Rolls 1 - 6 (controls) were not inoculated. The seedlings in Rolls 7 - 12 (treatments) were inoculated with nematodes in a 1% solution of methyl cellulose.

(ii) Rolls 13 - 24 contained seedlings of the lucerne variety Wairau. The seedlings in Rolls 13 - 18 (controls) were not inoculated. The seedlings in Rolls 19 - 24 (treatments) were inoculated with nematodes in a 1% solution of methyl cellulose.

(iii) Rolls 25 - 36 contained seedlings of the lucerne variety Grimm. The seedlings in Rolls 25 - 30 (controls) were not inoculated. The seedlings in Rolls 31 - 36 (treatments) were inoculated with nematodes in a 1% solution of methyl cellulose.

All seedlings in this experiment were grown for a period of three weeks in a growth cabinet at 16°C. The results of this experiment are given in Table 11. Assessment was by measurement of the stem diameters and the measurements for the whole sample are given in Appendix 1.
TABLE 11

RESULTS FROM THREE LUCERNE VARIETIES TESTED FOR NEMATODE SUSCEPTIBILITY

<table>
<thead>
<tr>
<th></th>
<th>Initial No. of Plants</th>
<th>Plants Surviving</th>
<th>Mortality</th>
<th>Mean Stem Diameter in mm.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Washoe</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Controls</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 1-3</td>
<td>45</td>
<td>42</td>
<td>3</td>
<td>2.75</td>
</tr>
<tr>
<td>Rolls 4-6</td>
<td>45</td>
<td>41</td>
<td>4</td>
<td>2.88</td>
</tr>
<tr>
<td><strong>Treatments</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 7-9</td>
<td>45</td>
<td>40</td>
<td>5</td>
<td>4.67</td>
</tr>
<tr>
<td>Rolls 10-12</td>
<td>45</td>
<td>39</td>
<td>6</td>
<td>4.58</td>
</tr>
<tr>
<td><strong>Totals:</strong></td>
<td>180</td>
<td>162</td>
<td>18</td>
<td></td>
</tr>
<tr>
<td><strong>Wairau</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Controls</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 13-15</td>
<td>45</td>
<td>40</td>
<td>5</td>
<td>2.83</td>
</tr>
<tr>
<td>Rolls 16-18</td>
<td>45</td>
<td>43</td>
<td>2</td>
<td>2.89</td>
</tr>
<tr>
<td><strong>Treatments</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 19-21</td>
<td>45</td>
<td>40</td>
<td>5</td>
<td>5.74</td>
</tr>
<tr>
<td>Rolls 22-24</td>
<td>45</td>
<td>41</td>
<td>4</td>
<td>5.52</td>
</tr>
<tr>
<td><strong>Totals:</strong></td>
<td>180</td>
<td>164</td>
<td>16</td>
<td></td>
</tr>
</tbody>
</table>

continued/...
### TABLE 11 (Continued)

<table>
<thead>
<tr>
<th>Initial No. of Plants</th>
<th>Plants Surviving</th>
<th>Mortality</th>
<th>Mean Stem Diameter in mm.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Grimm</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Controls</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 25-27</td>
<td>45</td>
<td>40</td>
<td>5</td>
</tr>
<tr>
<td>Rolls 28-30</td>
<td>45</td>
<td>38</td>
<td>7</td>
</tr>
<tr>
<td><strong>Treatments</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 31-33</td>
<td>45</td>
<td>41</td>
<td>4</td>
</tr>
<tr>
<td>Rolls 34-36</td>
<td>45</td>
<td>39</td>
<td>6</td>
</tr>
<tr>
<td><strong>Totals:</strong> 180</td>
<td>158</td>
<td>22</td>
<td></td>
</tr>
</tbody>
</table>

A sample of the measured plants from each of the three varieties was macerated with water and examined to see whether there were nematodes (larvae and adults bulked) and eggs present. The means of this sample are given in Table 12.
### TABLE 12
RESULTS FROM THE MACERATED LUCERNE SEEDLINGS FROM EXPERIMENT 12

<table>
<thead>
<tr>
<th>Variety</th>
<th>Washoe</th>
<th>Wairau</th>
<th>Grimm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of plants tested</td>
<td>77</td>
<td>80</td>
<td>81</td>
</tr>
<tr>
<td>Number of plants with nematodes and eggs</td>
<td>32</td>
<td>47</td>
<td>52</td>
</tr>
<tr>
<td>Average stem diameter of plants with nematodes and eggs (in mm.)</td>
<td>5.81</td>
<td>6.52</td>
<td>6.79</td>
</tr>
<tr>
<td>Number of plants with nematodes only</td>
<td>29</td>
<td>24</td>
<td>21</td>
</tr>
<tr>
<td>Average stem diameter of plants with nematodes only (in mm.)</td>
<td>4.25</td>
<td>4.82</td>
<td>4.74</td>
</tr>
<tr>
<td>Plants with no attack at all</td>
<td>16</td>
<td>9</td>
<td>8</td>
</tr>
<tr>
<td>Average stem diameter of plants with no attack (in mm.)</td>
<td>3.19</td>
<td>3.09</td>
<td>3.38</td>
</tr>
</tbody>
</table>

If the ability of the nematodes to reproduce within a plant is a sign that the plant is susceptible to attack, then the percentage of plants containing reproducing nematodes should give an indication of the susceptibility of the variety.

These percentages are as follows:

- Washoe 41.6%
- Wairau 58.8%
- Grimm 64.2%
Experiment 13

Fifty-four filter paper rolls with 15 lucerne seedlings in each were set up as follows: (A unit was taken as 45 plants and thus made up of three rolls.)

(i) Rolls 1 - 18 contained seedlings of the lucerne variety Washoe. The seedlings in Rolls 1 - 6 (controls) were not inoculated. The seedlings in Rolls 7 - 18 (treatments) were inoculated with nematodes in a 1% solution of methyl cellulose.

(ii) Rolls 19 - 36 contained seedlings of the lucerne variety Wairau. The seedlings in Rolls 19 - 24 (controls) were not inoculated. The seedlings in Rolls 25 - 36 (treatments) were inoculated with nematodes in a 1% solution of methyl cellulose.

(iii) Rolls 37 - 54 contained seedlings of the lucerne variety Grimm. The seedlings in Rolls 37 - 42 (controls) were not inoculated. The seedlings in Rolls 43 - 54 (treatments) were inoculated with nematodes in a 1% solution of methyl cellulose.

All seedlings in this experiment were grown for three weeks in a growth cabinet at 16°C. The results of this experiment are given in Table 13. Assessment was by measurement of the stem diameters, and the measurements for the whole sample are given in Appendix 1.
## TABLE 13

### RESULTS FROM THREE LUCERNE VARIETIES

**TESTED FOR NEMATODE SUSCEPTIBILITY**

<table>
<thead>
<tr>
<th>Initial No. of Plants</th>
<th>Plants Surviving</th>
<th>Mortality</th>
<th>Mean Stem Diameter in mm.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Washoe</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Controls</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 1–3</td>
<td>45</td>
<td>39</td>
<td>6</td>
</tr>
<tr>
<td>Rolls 4–6</td>
<td>45</td>
<td>39</td>
<td>6</td>
</tr>
<tr>
<td><strong>Treatments</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 7–9</td>
<td>45</td>
<td>38</td>
<td>7</td>
</tr>
<tr>
<td>Rolls 10–12</td>
<td>45</td>
<td>36</td>
<td>9</td>
</tr>
<tr>
<td>Rolls 13–15</td>
<td>45</td>
<td>37</td>
<td>8</td>
</tr>
<tr>
<td>Rolls 16–18</td>
<td>45</td>
<td>37</td>
<td>8</td>
</tr>
<tr>
<td><strong>Totals:</strong></td>
<td>270</td>
<td>226</td>
<td>44</td>
</tr>
<tr>
<td><strong>Wairau</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Controls</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 19–21</td>
<td>45</td>
<td>41</td>
<td>4</td>
</tr>
<tr>
<td>Rolls 22–24</td>
<td>45</td>
<td>43</td>
<td>2</td>
</tr>
<tr>
<td><strong>Treatments</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rolls 25–27</td>
<td>45</td>
<td>35</td>
<td>10</td>
</tr>
<tr>
<td>Rolls 28–30</td>
<td>45</td>
<td>44</td>
<td>1</td>
</tr>
<tr>
<td>Rolls 31–33</td>
<td>45</td>
<td>40</td>
<td>5</td>
</tr>
<tr>
<td>Rolls 34–36</td>
<td>45</td>
<td>38</td>
<td>7</td>
</tr>
</tbody>
</table>
A sample of the measured plants from each of the three varieties was macerated with water and examined to see whether there were nematodes (larvae and adults bulked) and eggs present. The means of this sample are given in Table 14.
TABLE 14

RESULTS FROM THE MACERATED LUCERNE SEEDLINGS FROM EXPERIMENT 13

<table>
<thead>
<tr>
<th>Variety</th>
<th>Washoe</th>
<th>Wairau</th>
<th>Grimm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of plants tested</td>
<td>72</td>
<td>78</td>
<td>76</td>
</tr>
<tr>
<td>Number of plants with nematodes and eggs</td>
<td>28</td>
<td>45</td>
<td>47</td>
</tr>
<tr>
<td>Average stem diameter of plants with nematodes and eggs (in mm.)</td>
<td>5.23</td>
<td>5.84</td>
<td>5.94</td>
</tr>
<tr>
<td>Number of plants with nematodes only</td>
<td>26</td>
<td>19</td>
<td>20</td>
</tr>
<tr>
<td>Average stem diameter of plants with nematodes only (in mm.)</td>
<td>4.19</td>
<td>3.95</td>
<td>4.70</td>
</tr>
<tr>
<td>Plants with no attack at all</td>
<td>17</td>
<td>15</td>
<td>9</td>
</tr>
<tr>
<td>Average stem diameter of plants with no attack (in mm.)</td>
<td>3.18</td>
<td>2.97</td>
<td>3.36</td>
</tr>
</tbody>
</table>

If the ability of the nematodes to reproduce within a plant is a sign that the plant is susceptible to attack, then the percentage of plants containing reproducing nematodes should give an indication of the susceptibility of the variety.

These percentages are as follows:

- Washoe 38.9%
- Wairau 57.6%
- Grimm 61.8%
The means of the stem diameters for all seedlings in Experiments 11, 12 and 13 are given in Table 15.

**Table 15**

<table>
<thead>
<tr>
<th>Treatment &amp; Variety</th>
<th>Washoe</th>
<th>Wairau</th>
<th>Grimm</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Experiment 11</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Controls</td>
<td>2.72</td>
<td>2.96</td>
<td>2.98</td>
</tr>
<tr>
<td></td>
<td>2.83</td>
<td>3.02</td>
<td>3.03</td>
</tr>
<tr>
<td>Inoc.</td>
<td>3.98</td>
<td>4.53</td>
<td>5.21</td>
</tr>
<tr>
<td></td>
<td>3.76</td>
<td>4.58</td>
<td>5.40</td>
</tr>
<tr>
<td></td>
<td>3.88</td>
<td>4.57</td>
<td>4.97</td>
</tr>
<tr>
<td></td>
<td>3.79</td>
<td>4.77</td>
<td>5.38</td>
</tr>
<tr>
<td><strong>Experiment 12</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Controls</td>
<td>2.75</td>
<td>2.83</td>
<td>2.90</td>
</tr>
<tr>
<td></td>
<td>2.88</td>
<td>2.89</td>
<td>2.99</td>
</tr>
<tr>
<td>Inoc.</td>
<td>4.67</td>
<td>5.74</td>
<td>5.96</td>
</tr>
<tr>
<td></td>
<td>4.58</td>
<td>5.52</td>
<td>5.89</td>
</tr>
<tr>
<td><strong>Experiment 13</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Controls</td>
<td>2.75</td>
<td>2.78</td>
<td>2.91</td>
</tr>
<tr>
<td></td>
<td>2.78</td>
<td>2.69</td>
<td>2.94</td>
</tr>
<tr>
<td>Inoc.</td>
<td>4.34</td>
<td>4.92</td>
<td>5.41</td>
</tr>
<tr>
<td></td>
<td>4.38</td>
<td>4.82</td>
<td>5.27</td>
</tr>
<tr>
<td></td>
<td>4.25</td>
<td>5.05</td>
<td>5.37</td>
</tr>
<tr>
<td></td>
<td>4.32</td>
<td>4.95</td>
<td>5.45</td>
</tr>
</tbody>
</table>
The data in Table 15 was transformed to $\log_{10}$ of the mean stem diameter as shown in Table 16.

### TABLE 16

**TRANSFORMED DATA FROM TABLE 15 - (LOG 10 OF MEAN STEM DIAMETER)**

<table>
<thead>
<tr>
<th>Treatment &amp; Variety</th>
<th>Washoe</th>
<th>Weirau</th>
<th>Grimm</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Experiment 11</strong></td>
<td>Controls</td>
<td>.435</td>
<td>.471</td>
</tr>
<tr>
<td></td>
<td></td>
<td>.452</td>
<td>.481</td>
</tr>
<tr>
<td></td>
<td>Subtotal</td>
<td>.887</td>
<td>.952</td>
</tr>
<tr>
<td></td>
<td>Inoculations</td>
<td>.600</td>
<td>.656</td>
</tr>
<tr>
<td></td>
<td></td>
<td>.575</td>
<td>.661</td>
</tr>
<tr>
<td></td>
<td></td>
<td>.589</td>
<td>.660</td>
</tr>
<tr>
<td></td>
<td></td>
<td>.759</td>
<td>.678</td>
</tr>
<tr>
<td></td>
<td>Subtotal</td>
<td>2.343</td>
<td>2.655</td>
</tr>
<tr>
<td></td>
<td>Total:</td>
<td>3.230</td>
<td>3.607</td>
</tr>
<tr>
<td><strong>Experiment 12</strong></td>
<td>Controls</td>
<td>.439</td>
<td>.452</td>
</tr>
<tr>
<td></td>
<td></td>
<td>.459</td>
<td>.462</td>
</tr>
<tr>
<td></td>
<td>Subtotal</td>
<td>.898</td>
<td>.914</td>
</tr>
<tr>
<td></td>
<td>Inoculations</td>
<td>.669</td>
<td>.759</td>
</tr>
<tr>
<td></td>
<td></td>
<td>.661</td>
<td>.742</td>
</tr>
<tr>
<td></td>
<td>Subtotal</td>
<td>1.330</td>
<td>1.501</td>
</tr>
<tr>
<td></td>
<td>Total:</td>
<td>2.228</td>
<td>2.415</td>
</tr>
</tbody>
</table>

continued/...
An analysis of variance following Sokal and Rohlf (1969) was then carried out and this is shown in Table 17.

**TABLE 17**

<table>
<thead>
<tr>
<th>Source</th>
<th>df</th>
<th>ss</th>
<th>ms</th>
<th>F</th>
</tr>
</thead>
<tbody>
<tr>
<td>Among experiments</td>
<td>2</td>
<td>.002202</td>
<td>.001101</td>
<td>1.75 N.S.</td>
</tr>
<tr>
<td>Among varieties</td>
<td>2</td>
<td>.052064</td>
<td>.026032</td>
<td>27.3***</td>
</tr>
<tr>
<td>Between treatments</td>
<td>1</td>
<td>.57.952</td>
<td>.571952</td>
<td>599.3***</td>
</tr>
<tr>
<td>Error</td>
<td>42</td>
<td>.04.0054</td>
<td>.000954</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>47</td>
<td>.666272</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
A further analysis of variance (Sokal and Rohlf, 1969) was carried out on the mean stem diameters of the plants in Experiments 11, 12 and 13 that contained nematodes and eggs. The numbers of plants infected are listed in Tables 10, 12 and 14. The stem diameters of these plants are included in Appendix 2. The analysis of variance of these is shown in Table 18.

**TABLE 18**

**ANALYSIS OF VARIANCE OF PLANTS CONTAINING NEMATODES AND EGGS**

<table>
<thead>
<tr>
<th>Source</th>
<th>df</th>
<th>ss</th>
<th>ms</th>
<th>F</th>
</tr>
</thead>
<tbody>
<tr>
<td>Among varieties</td>
<td>2</td>
<td>44.49</td>
<td>22.25</td>
<td>22.03***</td>
</tr>
<tr>
<td>Error</td>
<td>371</td>
<td>375.92</td>
<td>1.01</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>373</td>
<td>420.41</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

The percentage of plants that had reproducing nematodes in them when they were macerated in Experiments 11, 12 and 13 is shown in Table 19.

**TABLE 19**

**PERCENTAGE OF PLANTS CONTAINING REPRODUCING NEMATODES**

<table>
<thead>
<tr>
<th>Lucerne Variety</th>
<th>Experiment 11</th>
<th>Experiment 12</th>
<th>Experiment 13</th>
</tr>
</thead>
<tbody>
<tr>
<td>Washoe</td>
<td>9.6</td>
<td>41.6</td>
<td>38.9</td>
</tr>
<tr>
<td>Wairau</td>
<td>53.4</td>
<td>58.8</td>
<td>57.6</td>
</tr>
<tr>
<td>Grimm</td>
<td>52.9</td>
<td>64.2</td>
<td>61.8</td>
</tr>
</tbody>
</table>
In Experiment 13 a subsample was taken and a score on the following basis was given to the number of nematodes. This was with 30 plants of the three varieties, Washoe, Wairau and Grimm. The system used was that of Bingefors (1957), (pp. 15)

\[
\begin{align*}
0 &= \text{No nematodes or eggs} \\
1 &= \text{Nematodes but no eggs} \\
2 &= \text{A few nematodes and eggs} \\
3 &= \text{Many nematodes and eggs}
\end{align*}
\]

An index of resistance can be calculated from the following formula:

\[
\text{Index of resistance} = \left(1 - \frac{\sum a}{3n}\right) \times 100
\]

where \(a\) is the classification of each individual plant, and \(n\) is the number of observed plants.

Using the results tabulated in Table 20, the following indexes were obtained:

- **Washoe**: 48
- **Wairau**: 28
- **Grimm**: 26
TABLE 20

COMPARISON OF STEM DIAMETER OF SEEDLINGS WITH RATINGS OF NEMATODE NUMBERS AND REPRODUCTION WITHIN THE PLANTS FOR 30 PLANTS FROM EXPERIMENT 13

<table>
<thead>
<tr>
<th>Washoe Diameter of plant in mm.</th>
<th>Rating</th>
<th>Weirau Diameter of plant in mm.</th>
<th>Rating</th>
<th>Grimm Diameter of plant in mm.</th>
<th>Rating</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>4.2</td>
<td>2</td>
<td>3.8</td>
<td>0</td>
<td>7.0</td>
</tr>
<tr>
<td>2</td>
<td>5.0</td>
<td>3</td>
<td>6.4</td>
<td>0</td>
<td>6.8</td>
</tr>
<tr>
<td>3</td>
<td>3.3</td>
<td>2</td>
<td>5.4</td>
<td>3</td>
<td>6.3</td>
</tr>
<tr>
<td>4</td>
<td>5.2</td>
<td>2</td>
<td>5.3</td>
<td>3</td>
<td>3.5</td>
</tr>
<tr>
<td>5</td>
<td>4.2</td>
<td>1</td>
<td>6.0</td>
<td>3</td>
<td>3.3</td>
</tr>
<tr>
<td>6</td>
<td>3.6</td>
<td>1</td>
<td>4.5</td>
<td>3</td>
<td>6.8</td>
</tr>
<tr>
<td>7</td>
<td>5.2</td>
<td>3</td>
<td>4.8</td>
<td>2</td>
<td>6.5</td>
</tr>
<tr>
<td>8</td>
<td>5.2</td>
<td>2</td>
<td>5.8</td>
<td>3</td>
<td>4.6</td>
</tr>
<tr>
<td>9</td>
<td>3.8</td>
<td>1</td>
<td>3.8</td>
<td>2</td>
<td>4.3</td>
</tr>
<tr>
<td>10</td>
<td>2.3</td>
<td>0</td>
<td>5.0</td>
<td>3</td>
<td>6.3</td>
</tr>
<tr>
<td>11</td>
<td>3.6</td>
<td>0</td>
<td>4.0</td>
<td>3</td>
<td>7.0</td>
</tr>
<tr>
<td>12</td>
<td>4.2</td>
<td>2</td>
<td>4.0</td>
<td>1</td>
<td>6.7</td>
</tr>
<tr>
<td>13</td>
<td>7.3</td>
<td>3</td>
<td>4.8</td>
<td>1</td>
<td>7.2</td>
</tr>
<tr>
<td>14</td>
<td>5.8</td>
<td>2</td>
<td>3.0</td>
<td>1</td>
<td>6.8</td>
</tr>
<tr>
<td>15</td>
<td>5.2</td>
<td>2</td>
<td>5.8</td>
<td>3</td>
<td>5.5</td>
</tr>
<tr>
<td>16</td>
<td>3.6</td>
<td>2</td>
<td>3.3</td>
<td>1</td>
<td>7.0</td>
</tr>
<tr>
<td>17</td>
<td>6.2</td>
<td>3</td>
<td>6.4</td>
<td>3</td>
<td>4.5</td>
</tr>
<tr>
<td>18</td>
<td>5.8</td>
<td>3</td>
<td>7.0</td>
<td>3</td>
<td>4.0</td>
</tr>
<tr>
<td>19</td>
<td>5.0</td>
<td>1</td>
<td>6.0</td>
<td>3</td>
<td>4.2</td>
</tr>
<tr>
<td>20</td>
<td>5.2</td>
<td>3</td>
<td>7.0</td>
<td>3</td>
<td>4.0</td>
</tr>
<tr>
<td>21</td>
<td>5.6</td>
<td>1</td>
<td>6.3</td>
<td>3</td>
<td>6.3</td>
</tr>
<tr>
<td>22</td>
<td>2.7</td>
<td>0</td>
<td>5.5</td>
<td>2</td>
<td>4.0</td>
</tr>
<tr>
<td>23</td>
<td>3.1</td>
<td>0</td>
<td>7.8</td>
<td>3</td>
<td>4.0</td>
</tr>
<tr>
<td>24</td>
<td>4.0</td>
<td>1</td>
<td>2.8</td>
<td>1</td>
<td>5.4</td>
</tr>
<tr>
<td>25</td>
<td>3.2</td>
<td>0</td>
<td>7.8</td>
<td>3</td>
<td>5.8</td>
</tr>
<tr>
<td>26</td>
<td>6.4</td>
<td>2</td>
<td>7.2</td>
<td>3</td>
<td>5.2</td>
</tr>
<tr>
<td>27</td>
<td>5.1</td>
<td>1</td>
<td>6.8</td>
<td>3</td>
<td>5.0</td>
</tr>
<tr>
<td>28</td>
<td>3.8</td>
<td>1</td>
<td>3.5</td>
<td>0</td>
<td>6.8</td>
</tr>
<tr>
<td>29</td>
<td>3.6</td>
<td>0</td>
<td>3.8</td>
<td>1</td>
<td>6.3</td>
</tr>
<tr>
<td>30</td>
<td>3.6</td>
<td>0</td>
<td>3.1</td>
<td>0</td>
<td>4.6</td>
</tr>
</tbody>
</table>
Initially in this research project, time was devoted to perfecting a technique for growing lucerne seedlings under conditions in which they could be tested for resistance to the stem nematode *Ditylenchus dipsaci*. Two ways of growing seedlings were tried. These were, growing the seedlings in filter paper rolls, a technique similar to that used by earlier workers (Bingefors, 1957; Dijkstra, 1956; Frandsen, 1951; Grundbacher, 1962; Hanna and Hawn, 1965), or, growing the seedlings in soil flats. Work was originally concentrated on the soil flats because of disease problems that occurred in initial attempts to grow the seedlings in filter paper rolls. Eventually these disease problems were eliminated and the filter paper roll technique was used in preference to the soil flat technique for the following reasons. The filter paper rolls were more convenient to handle and work with than the soil flats; many more plants could be dealt with; the environment could be controlled more accurately by the use of growth cabinets instead of having the plants in a glasshouse as was necessary with the soil flats; the time period for each experiment was shortened. Assessment of the plants in soil flats was carried out on a visual basis, swollen plants being classified as susceptible. The results of these assessments, which were carried out two, three and four weeks after inoculation, are given in Tables 1, 2 and 3. There was no uniformity of infection obtained between replications 1, 2, 3 and 4 in this experiment, but it is possible to say that about 65% of the 800 seedlings inoculated became infected. The fact that uniformity
of infection between replications in this experiment could not be obtained was another important reason for the rejection of the soil flat technique by the author. The plants that were inoculated with a 1% solution of methyl cellulose only in the soil flat trials showed no swelling, so that it is possible to say that it was definitely the nematodes that caused the swelling in the plants inoculated with nematodes in a 1% solution of methyl cellulose.

The results for the work using the filter paper roll technique are listed in Chapter IV in a sequence of experiments from 1 - 13. The findings for Experiment 1 are given in Table 4 and it can be seen that the mortality of seedlings due to fungal attack was so high that no worthwhile results on resistance of the lucerne could be obtained. Experiments 2 - 8 were all failures due to fungal attack, which was found to have originated from a heavily infected seed line. This line was rejected before Experiment 9. However, during these experiments many useful points of technique evolved. It was found that 50 plants per roll were too many because the paper size used in the rolls (11.5 cm. x 57 cm.) was too large to work with efficiently. As well as this the 3% solution of methyl cellulose caused the plants to stick together and also caused the binding of the apical regions, thus inhibiting growth. The paper size for the rolls was subsequently reduced to 11.5 cm. x 22.5 cm., the number of seedlings per roll reduced to 15 and a 1% solution of methyl cellulose was used instead of the 3% solution.

Throughout all these experiments an average of 50 nematodes per drop of inoculum was used and each plant was inoculated twice.

In Experiment 9, 15 seedlings were used in rolls made from filter papers of the new dimensions. The plants were inoculated and grown
successfully for three weeks. The symptoms were visually assessed and the results are given in Tables 5, 6 and 7. Fungal disease was not a problem in this experiment because the seed line had been changed and an efficient method for the surface sterilization of the seed was being used. In this experiment there was no swelling in the plants that were inoculated with a methyl cellulose solution only, so again it can be said that swelling in the plants in the filter paper rolls was due to the nematodes. This experiment brought to an end the phase of work on the technique of growing seedlings in filter paper rolls. Up to this stage all the work had been carried out with Wairau lucerne.

In Experiment 10 three varieties of lucerne were grown in filter paper rolls. These were Wairau, the relative resistance of which is unknown, Lahontan and Washoe, which are varieties that have been bred in North America for resistance to stem nematode attack. The results of a visual assessment are given in Table 8. This experiment showed that it was quite possible to grow lucerne varieties other than Wairau in filter paper rolls.

Experiments 11, 12 and 13 were designed to compare three different varieties of lucerne, Washoe, Wairau and Grimm, for their resistance to stem nematode. Washoe has been bred for resistance to nematode attack, Wairau has an unknown relative resistance, while Grimm is known to be susceptible. In these three experiments the system of rating the plants on visual symptoms was improved upon by using an eyepiece graticule in a stereoscopic microscope to measure the stem diameters. The plant mortalities and the average stem diameters are given in Tables 9, 11 and 13 for Experiments 11, 12 and 13 respectively. A sample of plants of each variety from every experiment was macerated with water, examined under a
stereoscopic microscope and split into three categories as follows: those with nematodes and eggs present, those with nematodes only present and those with neither nematodes nor eggs present. Tables 10, 12 and 14 express the proportions of the samples in each category and also give the means of the sample.

Table 15 gives the mean stem diameters in mm. of 45 seedlings of three varieties of lucerne in three experiments, and Table 16 shows these figures transformed to log $\log_{10}$.

An analysis of variance of the transformed means of the stem diameters (Table 17) showed that there were highly significant differences between the varieties in the degree of swelling due to nematode attack. Grimm showed the greatest swelling, Washoe the least, while Wairau was intermediate. There were also highly significant differences between control and treated plants which was expected. This analysis showed no significant differences between the experiments.

An analysis of variance was carried out on the diameters of those plants in Experiments 11, 12 and 13 that did have nematode reproduction occurring. Again, highly significant differences were obtained between varieties, as shown in Table 18.

Table 19 is a summary table of the percentages of plants in which nematode reproduction occurred in the three Experiments 11, 12 and 13. From these figures it can be seen that the percentages of the plants tested that had reproducing nematodes were very similar for Wairau and Grimm, but were much lower for Washoe. The fact that the percentages of plants infected in Experiment 11 were generally lower, with Washoe being much lower, can be explained by the fact that this was the first experiment where testing for reproduction was carried out and the technique of scanning
for eggs and nematodes had not been perfected. Table 20 shows that the ratings given to Washoe seedlings are, in general, lower than the ratings given to Wairau and Grimm seedlings. This also helps explain why the percentages of Washoe seedlings with nematode reproduction are so much lower in Experiment 11.

The method of obtaining a resistance rating for a variety of red clover, as used by Bingefors (1957), was used on a sample of 30 plants of each variety in Experiment 13. The ratings obtained placed Wairau and Grimm very close in their susceptibility, while Washoe was much more resistant. This rating agrees with the figures for the percentage of plants in each variety in which nematode reproduction occurred in Experiments 12 and 13, where Washoe was found to be more resistant than Grimm and Wairau, which were close together.

From these figures it can be seen that swelling alone cannot be used to give the relative resistance of different lucerne varieties, but from the results shown in Tables 10, 12 and 14 it can be seen that selections could be made within a variety on the basis of swelling. The differences between the average stem diameters of the control seedlings and those that were recorded as having no nematodes or eggs when they were tested, could be due to the fact that nematodes may have entered the seedlings and though they did not survive, nevertheless caused some slight swelling.

Comparing resistance of lucerne varieties to *Ditylenchus dipsaci* attack would be best carried out on the basis of the percentage of plants in each variety that contain reproducing nematodes. On this basis Wairau must be classified as susceptible.
CHAPTER VI

SUMMARY

The results of testing lucerne for resistance to the stem nematode *Ditylenchus dipsaci* are recorded. A major part of the study was given to perfecting a technique for growing lucerne seedlings for inoculation with nematodes. Two techniques were tried and filter paper rolls were found to be superior to soil flats.

The technique of growing seedlings in filter paper rolls and inoculating them with nematodes was used to produce a rating of the resistance for the New Zealand variety of lucerne, Wairau, when compared with a known resistant variety, Washoe and a susceptible variety, Grimm. From the results obtained Wairau must be classified as a susceptible variety.

It appears that a selection within a variety of lucerne for resistance to *Ditylenchus dipsaci* could be made by eliminating the swollen plants. However, to obtain the relative resistances of several varieties, it would be necessary to obtain a measure of the reproductive rates of the nematodes in the plants.
ACKNOWLEDGEMENTS

I wish to thank Dr. C.P. Hoyt for his supervision during this project and his advice on the preparation of this manuscript. I also wish to thank Professor R.A. Harrison for his valuable assistance during this work.

The co-operation of Mr. A. Wallace during the statistical analysis is gratefully acknowledged and appreciation is expressed to Mr. J. Miles for his help with the photography.

Thanks are due to Mr. Rex Copland and Mr. A.T. Metherell for allowing me to collect nematode infected lucerne from their properties.

Financial assistance from the N.Z. Wool Board and the D.S.I.R. is gratefully acknowledged.

Finally I wish to thank Mr. D. Ives for proof reading and Mrs. V. Judd for her very capable typing of the manuscript.
REFERENCES


GOODEY, T. 1929. The stem eelworm *Tylenchus dipsaci*; observations on its attack on potatoes and marigolds, with a host list of plants parasitized by it. *J. Helminth.* 7: 183-200.


# APPENDIX 1

## STEM MEASUREMENTS FOR INDIVIDUAL LUCERNE SEEDLINGS IN mm.

<table>
<thead>
<tr>
<th>Washoe Plant No.</th>
<th>Controls Rolls 1-3</th>
<th>Controls Rolls 4-6</th>
<th>Experiment 11 Rolls 7-9</th>
<th>Treatments Rolls 10-12</th>
<th>Treatments Rolls 13-15</th>
<th>Treatments Rolls 16-18</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2.3</td>
<td>2.3</td>
<td>3.4</td>
<td>4.5</td>
<td>3.1</td>
<td>3.5</td>
</tr>
<tr>
<td>2</td>
<td>2.2</td>
<td>4.0</td>
<td>6.0</td>
<td>3.5</td>
<td>3.3</td>
<td>5.0</td>
</tr>
<tr>
<td>3</td>
<td>2.6</td>
<td>3.3</td>
<td>4.2</td>
<td>2.2</td>
<td>3.4</td>
<td>3.4</td>
</tr>
<tr>
<td>4</td>
<td>3.0</td>
<td>3.3</td>
<td>2.7</td>
<td>2.3</td>
<td>4.0</td>
<td>4.0</td>
</tr>
<tr>
<td>5</td>
<td>3.0</td>
<td>3.5</td>
<td>3.5</td>
<td>3.5</td>
<td>2.9</td>
<td>2.6</td>
</tr>
<tr>
<td>6</td>
<td>2.5</td>
<td>2.4</td>
<td>4.0</td>
<td>4.0</td>
<td>3.1</td>
<td>6.2</td>
</tr>
<tr>
<td>7</td>
<td>2.2</td>
<td>2.5</td>
<td>7.5</td>
<td>3.8</td>
<td>4.2</td>
<td>5.2</td>
</tr>
<tr>
<td>8</td>
<td>3.0</td>
<td>2.6</td>
<td>3.5</td>
<td>4.5</td>
<td>5.0</td>
<td>3.4</td>
</tr>
<tr>
<td>9</td>
<td>3.5</td>
<td>2.6</td>
<td>3.5</td>
<td>3.3</td>
<td>4.2</td>
<td>2.1</td>
</tr>
<tr>
<td>10</td>
<td>2.5</td>
<td>2.8</td>
<td>3.8</td>
<td>2.1</td>
<td>5.3</td>
<td>5.8</td>
</tr>
<tr>
<td>11</td>
<td>3.0</td>
<td>3.2</td>
<td>3.0</td>
<td>5.0</td>
<td>4.1</td>
<td>3.0</td>
</tr>
<tr>
<td>12</td>
<td>3.4</td>
<td>2.9</td>
<td>5.0</td>
<td>2.2</td>
<td>5.1</td>
<td>4.0</td>
</tr>
<tr>
<td>13</td>
<td>2.6</td>
<td>3.2</td>
<td>4.5</td>
<td>6.0</td>
<td>3.6</td>
<td>3.5</td>
</tr>
<tr>
<td>14</td>
<td>2.6</td>
<td>3.0</td>
<td>3.5</td>
<td>4.0</td>
<td>4.5</td>
<td>2.9</td>
</tr>
<tr>
<td>15</td>
<td>2.1</td>
<td>3.1</td>
<td>5.5</td>
<td>2.1</td>
<td>5.6</td>
<td>3.8</td>
</tr>
<tr>
<td>16</td>
<td>2.5</td>
<td>2.5</td>
<td>3.5</td>
<td>5.0</td>
<td>4.3</td>
<td>5.3</td>
</tr>
<tr>
<td>17</td>
<td>2.0</td>
<td>2.5</td>
<td>4.5</td>
<td>5.0</td>
<td>5.4</td>
<td>3.2</td>
</tr>
<tr>
<td>18</td>
<td>2.5</td>
<td>2.8</td>
<td>5.4</td>
<td>3.8</td>
<td>3.5</td>
<td>3.3</td>
</tr>
<tr>
<td>19</td>
<td>2.4</td>
<td>2.0</td>
<td>4.3</td>
<td>3.2</td>
<td>3.9</td>
<td>5.4</td>
</tr>
<tr>
<td>20</td>
<td>3.0</td>
<td>3.0</td>
<td>4.8</td>
<td>3.1</td>
<td>4.2</td>
<td>3.8</td>
</tr>
<tr>
<td>21</td>
<td>2.0</td>
<td>2.6</td>
<td>3.2</td>
<td>2.8</td>
<td>4.6</td>
<td>3.2</td>
</tr>
<tr>
<td>22</td>
<td>3.3</td>
<td>2.4</td>
<td>2.5</td>
<td>3.3</td>
<td>4.2</td>
<td>4.6</td>
</tr>
<tr>
<td>23</td>
<td>3.0</td>
<td>2.5</td>
<td>2.1</td>
<td>3.5</td>
<td>2.6</td>
<td>3.4</td>
</tr>
<tr>
<td>24</td>
<td>3.5</td>
<td>2.9</td>
<td>3.5</td>
<td>4.5</td>
<td>2.8</td>
<td>3.5</td>
</tr>
<tr>
<td>25</td>
<td>2.6</td>
<td>3.1</td>
<td>6.0</td>
<td>4.0</td>
<td>3.6</td>
<td>3.1</td>
</tr>
<tr>
<td>26</td>
<td>3.6</td>
<td>2.6</td>
<td>5.5</td>
<td>5.0</td>
<td>6.3</td>
<td>4.5</td>
</tr>
<tr>
<td>27</td>
<td>3.5</td>
<td>3.0</td>
<td>6.0</td>
<td>3.8</td>
<td>3.7</td>
<td>2.8</td>
</tr>
<tr>
<td>28</td>
<td>2.0</td>
<td>3.2</td>
<td>2.6</td>
<td>5.1</td>
<td>3.5</td>
<td>3.4</td>
</tr>
<tr>
<td>29</td>
<td>2.5</td>
<td>2.6</td>
<td>3.2</td>
<td>2.5</td>
<td>2.6</td>
<td>1.9</td>
</tr>
<tr>
<td>30</td>
<td>3.1</td>
<td>3.0</td>
<td>4.3</td>
<td>4.3</td>
<td>3.4</td>
<td>3.4</td>
</tr>
<tr>
<td>31</td>
<td>2.5</td>
<td>2.2</td>
<td>2.7</td>
<td>2.5</td>
<td>3.2</td>
<td>3.2</td>
</tr>
<tr>
<td>32</td>
<td>2.6</td>
<td>2.8</td>
<td>2.3</td>
<td>3.5</td>
<td>3.2</td>
<td>3.2</td>
</tr>
<tr>
<td>33</td>
<td>3.5</td>
<td>3.0</td>
<td>4.0</td>
<td>7.0</td>
<td>4.5</td>
<td>3.5</td>
</tr>
<tr>
<td>34</td>
<td>2.0</td>
<td>3.0</td>
<td>3.0</td>
<td>3.2</td>
<td>4.1</td>
<td>4.1</td>
</tr>
<tr>
<td>35</td>
<td>2.5</td>
<td>3.0</td>
<td>3.0</td>
<td>2.5</td>
<td>3.8</td>
<td>4.9</td>
</tr>
<tr>
<td>36</td>
<td>2.8</td>
<td>4.3</td>
<td>4.3</td>
<td>4.3</td>
<td>4.3</td>
<td>4.3</td>
</tr>
<tr>
<td>Mean</td>
<td>2.77</td>
<td>2.92</td>
<td>3.09</td>
<td>3.16</td>
<td>3.55</td>
<td>3.56</td>
</tr>
</tbody>
</table>
**APPENDIX 1**

(Wairau Experiment)

(Continued)

<table>
<thead>
<tr>
<th>Plant No.</th>
<th>Controls (Rolls 19-21)</th>
<th>Controls (Rolls 22-24)</th>
<th>Treatments (Rolls 25-27)</th>
<th>Treatments (Rolls 28-30)</th>
<th>Treatments (Rolls 31-33)</th>
<th>Treatments (Rolls 34-36)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2.4</td>
<td>3.1</td>
<td>5.0</td>
<td>3.7</td>
<td>4.5</td>
<td>7.5</td>
</tr>
<tr>
<td>2</td>
<td>3.5</td>
<td>3.0</td>
<td>3.7</td>
<td>3.5</td>
<td>3.2</td>
<td>2.5</td>
</tr>
<tr>
<td>3</td>
<td>3.8</td>
<td>4.0</td>
<td>3.3</td>
<td>3.0</td>
<td>2.8</td>
<td>4.0</td>
</tr>
<tr>
<td>4</td>
<td>3.5</td>
<td>3.5</td>
<td>5.5</td>
<td>6.0</td>
<td>6.5</td>
<td>5.0</td>
</tr>
<tr>
<td>5</td>
<td>3.0</td>
<td>3.3</td>
<td>8.0</td>
<td>4.8</td>
<td>6.0</td>
<td>5.2</td>
</tr>
<tr>
<td>6</td>
<td>3.2</td>
<td>3.2</td>
<td>3.6</td>
<td>3.5</td>
<td>2.7</td>
<td>5.2</td>
</tr>
<tr>
<td>7</td>
<td>3.3</td>
<td>3.4</td>
<td>4.2</td>
<td>5.3</td>
<td>4.5</td>
<td>4.5</td>
</tr>
<tr>
<td>8</td>
<td>3.5</td>
<td>3.1</td>
<td>2.4</td>
<td>4.0</td>
<td>4.3</td>
<td>4.8</td>
</tr>
<tr>
<td>9</td>
<td>2.5</td>
<td>2.8</td>
<td>5.3</td>
<td>5.0</td>
<td>4.1</td>
<td>5.0</td>
</tr>
<tr>
<td>10</td>
<td>4.0</td>
<td>2.7</td>
<td>5.5</td>
<td>3.0</td>
<td>5.0</td>
<td>4.2</td>
</tr>
<tr>
<td>11</td>
<td>2.5</td>
<td>3.0</td>
<td>4.8</td>
<td>5.5</td>
<td>4.5</td>
<td>6.2</td>
</tr>
<tr>
<td>12</td>
<td>2.5</td>
<td>3.1</td>
<td>5.5</td>
<td>6.0</td>
<td>7.0</td>
<td>4.2</td>
</tr>
<tr>
<td>13</td>
<td>2.5</td>
<td>2.8</td>
<td>5.0</td>
<td>3.4</td>
<td>4.5</td>
<td>7.0</td>
</tr>
<tr>
<td>14</td>
<td>2.5</td>
<td>3.7</td>
<td>3.3</td>
<td>3.9</td>
<td>6.2</td>
<td>3.0</td>
</tr>
<tr>
<td>15</td>
<td>2.5</td>
<td>3.5</td>
<td>7.6</td>
<td>3.8</td>
<td>4.5</td>
<td>4.9</td>
</tr>
<tr>
<td>16</td>
<td>3.0</td>
<td>2.6</td>
<td>3.8</td>
<td>7.0</td>
<td>4.0</td>
<td>6.3</td>
</tr>
<tr>
<td>17</td>
<td>3.0</td>
<td>2.5</td>
<td>3.2</td>
<td>4.5</td>
<td>3.3</td>
<td>4.6</td>
</tr>
<tr>
<td>18</td>
<td>2.2</td>
<td>3.5</td>
<td>5.0</td>
<td>6.5</td>
<td>4.5</td>
<td>4.0</td>
</tr>
<tr>
<td>19</td>
<td>2.6</td>
<td>3.2</td>
<td>4.3</td>
<td>3.1</td>
<td>5.0</td>
<td>4.7</td>
</tr>
<tr>
<td>20</td>
<td>3.0</td>
<td>3.2</td>
<td>5.0</td>
<td>6.5</td>
<td>4.3</td>
<td>5.3</td>
</tr>
<tr>
<td>21</td>
<td>2.6</td>
<td>3.2</td>
<td>4.5</td>
<td>2.8</td>
<td>6.0</td>
<td>3.2</td>
</tr>
<tr>
<td>22</td>
<td>2.9</td>
<td>2.9</td>
<td>5.5</td>
<td>5.5</td>
<td>4.5</td>
<td>5.6</td>
</tr>
<tr>
<td>23</td>
<td>3.0</td>
<td>3.1</td>
<td>5.0</td>
<td>4.8</td>
<td>6.5</td>
<td>5.8</td>
</tr>
<tr>
<td>24</td>
<td>2.1</td>
<td>3.7</td>
<td>2.0</td>
<td>5.5</td>
<td>4.3</td>
<td>3.7</td>
</tr>
<tr>
<td>25</td>
<td>2.0</td>
<td>2.8</td>
<td>4.5</td>
<td>2.8</td>
<td>5.6</td>
<td>5.2</td>
</tr>
<tr>
<td>26</td>
<td>2.6</td>
<td>3.1</td>
<td>3.0</td>
<td>5.0</td>
<td>2.4</td>
<td>4.9</td>
</tr>
<tr>
<td>27</td>
<td>2.5</td>
<td>3.0</td>
<td>5.5</td>
<td>2.5</td>
<td>4.6</td>
<td>5.1</td>
</tr>
<tr>
<td>28</td>
<td>2.5</td>
<td>2.6</td>
<td>4.6</td>
<td>4.0</td>
<td>3.4</td>
<td>2.0</td>
</tr>
<tr>
<td>29</td>
<td>2.3</td>
<td>3.0</td>
<td>5.5</td>
<td>6.5</td>
<td>6.5</td>
<td>4.3</td>
</tr>
<tr>
<td>30</td>
<td>3.5</td>
<td>3.0</td>
<td>7.0</td>
<td>5.5</td>
<td>4.8</td>
<td>4.9</td>
</tr>
<tr>
<td>31</td>
<td>3.0</td>
<td>3.5</td>
<td>5.9</td>
<td>2.9</td>
<td>4.8</td>
<td>5.8</td>
</tr>
<tr>
<td>32</td>
<td>3.2</td>
<td>2.7</td>
<td>2.2</td>
<td>5.5</td>
<td>4.0</td>
<td>4.1</td>
</tr>
<tr>
<td>33</td>
<td>3.5</td>
<td>2.6</td>
<td>3.0</td>
<td>2.6</td>
<td>8.0</td>
<td>7.8</td>
</tr>
<tr>
<td>34</td>
<td>3.6</td>
<td>2.4</td>
<td>6.0</td>
<td>4.5</td>
<td>6.5</td>
<td>4.6</td>
</tr>
<tr>
<td>35</td>
<td>4.0</td>
<td>2.8</td>
<td>2.4</td>
<td>3.2</td>
<td>3.6</td>
<td>2.0</td>
</tr>
<tr>
<td>36</td>
<td>3.3</td>
<td>3.0</td>
<td>3.2</td>
<td>3.7</td>
<td>2.1</td>
<td>5.3</td>
</tr>
<tr>
<td>37</td>
<td>3.6</td>
<td>3.0</td>
<td>6.0</td>
<td>5.0</td>
<td>3.8</td>
<td>3.9</td>
</tr>
<tr>
<td>38</td>
<td>3.1</td>
<td>2.5</td>
<td>7.0</td>
<td>6.5</td>
<td>2.1</td>
<td>4.8</td>
</tr>
<tr>
<td>39</td>
<td>2.4</td>
<td>2.7</td>
<td>3.5</td>
<td>6.3</td>
<td>2.9</td>
<td></td>
</tr>
<tr>
<td>40</td>
<td>3.2</td>
<td>3.0</td>
<td>2.6</td>
<td>6.0</td>
<td>4.5</td>
<td></td>
</tr>
<tr>
<td>41</td>
<td>3.2</td>
<td>2.6</td>
<td>2.2</td>
<td>2.7</td>
<td>5.2</td>
<td></td>
</tr>
<tr>
<td>42</td>
<td>2.5</td>
<td>3.0</td>
<td>3.0</td>
<td>6.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>43</td>
<td>3.0</td>
<td>3.2</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>44</td>
<td>3.2</td>
<td>2.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>45</td>
<td>3.2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Mean: 2.96 3.02 4.53 4.58 4.57 4.77
## APPENDIX 1

### (Continued)

<table>
<thead>
<tr>
<th>Plant No.</th>
<th>Controls Rolls 37-39</th>
<th>Controls Rolls 40-42</th>
<th>Treatments Rolls 43-45</th>
<th>Treatments Rolls 46-48</th>
<th>Treatments Rolls 49-51</th>
<th>Treatments Rolls 52-54</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2.9</td>
<td>3.8</td>
<td>5.5</td>
<td>6.5</td>
<td>3.8</td>
<td>3.5</td>
</tr>
<tr>
<td>2</td>
<td>3.6</td>
<td>3.2</td>
<td>7.3</td>
<td>6.0</td>
<td>7.2</td>
<td>3.9</td>
</tr>
<tr>
<td>3</td>
<td>3.0</td>
<td>3.0</td>
<td>4.8</td>
<td>7.0</td>
<td>4.4</td>
<td>7.3</td>
</tr>
<tr>
<td>4</td>
<td>2.2</td>
<td>2.8</td>
<td>10.0</td>
<td>5.0</td>
<td>5.2</td>
<td>3.7</td>
</tr>
<tr>
<td>5</td>
<td>2.8</td>
<td>2.8</td>
<td>5.8</td>
<td>2.3</td>
<td>6.2</td>
<td>7.2</td>
</tr>
<tr>
<td>6</td>
<td>3.0</td>
<td>2.8</td>
<td>5.5</td>
<td>3.2</td>
<td>3.4</td>
<td>6.3</td>
</tr>
<tr>
<td>7</td>
<td>2.6</td>
<td>3.4</td>
<td>4.8</td>
<td>4.5</td>
<td>3.4</td>
<td>4.0</td>
</tr>
<tr>
<td>8</td>
<td>3.1</td>
<td>3.5</td>
<td>3.0</td>
<td>2.5</td>
<td>5.1</td>
<td>4.2</td>
</tr>
<tr>
<td>9</td>
<td>2.6</td>
<td>3.0</td>
<td>5.0</td>
<td>6.5</td>
<td>6.2</td>
<td>4.6</td>
</tr>
<tr>
<td>10</td>
<td>2.6</td>
<td>3.3</td>
<td>6.0</td>
<td>6.5</td>
<td>6.2</td>
<td>7.0</td>
</tr>
<tr>
<td>11</td>
<td>3.0</td>
<td>3.0</td>
<td>4.5</td>
<td>4.0</td>
<td>6.3</td>
<td>7.6</td>
</tr>
<tr>
<td>12</td>
<td>2.6</td>
<td>2.8</td>
<td>4.0</td>
<td>7.5</td>
<td>5.6</td>
<td>8.2</td>
</tr>
<tr>
<td>13</td>
<td>2.1</td>
<td>3.2</td>
<td>4.0</td>
<td>5.0</td>
<td>2.6</td>
<td>3.8</td>
</tr>
<tr>
<td>14</td>
<td>3.2</td>
<td>3.2</td>
<td>2.8</td>
<td>7.0</td>
<td>6.2</td>
<td>7.6</td>
</tr>
<tr>
<td>15</td>
<td>3.0</td>
<td>2.7</td>
<td>7.0</td>
<td>5.0</td>
<td>3.2</td>
<td>6.7</td>
</tr>
<tr>
<td>16</td>
<td>3.1</td>
<td>2.6</td>
<td>8.0</td>
<td>4.0</td>
<td>5.8</td>
<td>6.7</td>
</tr>
<tr>
<td>17</td>
<td>3.8</td>
<td>3.3</td>
<td>4.7</td>
<td>6.0</td>
<td>3.3</td>
<td>4.8</td>
</tr>
<tr>
<td>18</td>
<td>2.7</td>
<td>2.5</td>
<td>5.0</td>
<td>7.0</td>
<td>4.5</td>
<td>3.5</td>
</tr>
<tr>
<td>19</td>
<td>3.2</td>
<td>3.0</td>
<td>6.0</td>
<td>4.0</td>
<td>3.6</td>
<td>4.0</td>
</tr>
<tr>
<td>20</td>
<td>3.0</td>
<td>3.6</td>
<td>5.3</td>
<td>5.5</td>
<td>6.2</td>
<td>5.8</td>
</tr>
<tr>
<td>21</td>
<td>2.9</td>
<td>3.1</td>
<td>3.0</td>
<td>5.0</td>
<td>3.6</td>
<td>2.9</td>
</tr>
<tr>
<td>22</td>
<td>3.0</td>
<td>2.9</td>
<td>5.7</td>
<td>5.8</td>
<td>3.5</td>
<td>8.0</td>
</tr>
<tr>
<td>23</td>
<td>3.0</td>
<td>3.2</td>
<td>4.3</td>
<td>6.5</td>
<td>6.4</td>
<td>5.1</td>
</tr>
<tr>
<td>24</td>
<td>3.8</td>
<td>3.4</td>
<td>6.5</td>
<td>6.5</td>
<td>5.8</td>
<td>3.1</td>
</tr>
<tr>
<td>25</td>
<td>3.0</td>
<td>3.2</td>
<td>4.3</td>
<td>8.0</td>
<td>4.2</td>
<td>6.8</td>
</tr>
<tr>
<td>26</td>
<td>3.1</td>
<td>3.9</td>
<td>2.3</td>
<td>4.5</td>
<td>4.5</td>
<td>3.3</td>
</tr>
<tr>
<td>27</td>
<td>3.5</td>
<td>2.6</td>
<td>5.5</td>
<td>7.5</td>
<td>5.8</td>
<td>7.5</td>
</tr>
<tr>
<td>28</td>
<td>2.8</td>
<td>3.0</td>
<td>7.0</td>
<td>7.6</td>
<td>6.0</td>
<td></td>
</tr>
<tr>
<td>29</td>
<td>3.3</td>
<td>3.8</td>
<td>2.0</td>
<td>3.4</td>
<td>6.9</td>
<td></td>
</tr>
<tr>
<td>30</td>
<td>2.2</td>
<td>2.9</td>
<td>4.0</td>
<td>7.1</td>
<td>7.5</td>
<td></td>
</tr>
<tr>
<td>31</td>
<td>2.7</td>
<td>2.9</td>
<td>2.6</td>
<td>6.5</td>
<td>4.6</td>
<td></td>
</tr>
<tr>
<td>32</td>
<td>2.6</td>
<td>2.8</td>
<td>7.0</td>
<td>3.7</td>
<td>6.0</td>
<td></td>
</tr>
<tr>
<td>33</td>
<td>2.7</td>
<td>2.8</td>
<td>4.6</td>
<td>5.2</td>
<td>2.2</td>
<td></td>
</tr>
<tr>
<td>34</td>
<td>3.3</td>
<td>2.2</td>
<td>6.0</td>
<td>5.7</td>
<td>2.8</td>
<td></td>
</tr>
<tr>
<td>35</td>
<td>3.1</td>
<td>2.7</td>
<td>6.0</td>
<td>4.2</td>
<td>6.8</td>
<td></td>
</tr>
<tr>
<td>36</td>
<td>3.2</td>
<td>2.5</td>
<td>7.0</td>
<td>3.4</td>
<td></td>
<td></td>
</tr>
<tr>
<td>37</td>
<td>3.3</td>
<td>3.5</td>
<td>8.5</td>
<td>5.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>38</td>
<td>2.4</td>
<td>3.8</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>39</td>
<td>3.1</td>
<td>3.8</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>40</td>
<td>3.1</td>
<td>3.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>41</td>
<td></td>
<td>6.0</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>42</td>
<td></td>
<td>7.0</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>43</td>
<td></td>
<td>3.8</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>44</td>
<td></td>
<td>3.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>45</td>
<td></td>
<td>6.0</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Mean:** 2.98 3.03 5.21 5.40 4.97 5.38
## APPENDIX 1

(Continued)

<table>
<thead>
<tr>
<th>Plant No.</th>
<th>Control Rolls 1-3</th>
<th>Control Rolls 4-6</th>
<th>Treatment Rolls 7-9</th>
<th>Treatment Rolls 10-12</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2.9</td>
<td>2.3</td>
<td>4.0</td>
<td>4.5</td>
</tr>
<tr>
<td>2</td>
<td>3.1</td>
<td>4.0</td>
<td>7.8</td>
<td>7.3</td>
</tr>
<tr>
<td>3</td>
<td>3.5</td>
<td>3.3</td>
<td>3.5</td>
<td>3.1</td>
</tr>
<tr>
<td>4</td>
<td>2.6</td>
<td>3.3</td>
<td>3.1</td>
<td>2.5</td>
</tr>
<tr>
<td>5</td>
<td>2.2</td>
<td>3.5</td>
<td>5.6</td>
<td>4.6</td>
</tr>
<tr>
<td>6</td>
<td>3.4</td>
<td>2.4</td>
<td>5.3</td>
<td>4.5</td>
</tr>
<tr>
<td>7</td>
<td>2.7</td>
<td>2.5</td>
<td>4.5</td>
<td>4.2</td>
</tr>
<tr>
<td>8</td>
<td>2.9</td>
<td>2.6</td>
<td>4.1</td>
<td>6.0</td>
</tr>
<tr>
<td>9</td>
<td>3.0</td>
<td>3.6</td>
<td>6.0</td>
<td>2.7</td>
</tr>
<tr>
<td>10</td>
<td>3.0</td>
<td>2.8</td>
<td>3.5</td>
<td>4.0</td>
</tr>
<tr>
<td>11</td>
<td>2.9</td>
<td>3.2</td>
<td>4.0</td>
<td>3.0</td>
</tr>
<tr>
<td>12</td>
<td>3.0</td>
<td>3.4</td>
<td>3.0</td>
<td>2.8</td>
</tr>
<tr>
<td>13</td>
<td>2.4</td>
<td>2.9</td>
<td>3.0</td>
<td>6.5</td>
</tr>
<tr>
<td>14</td>
<td>2.6</td>
<td>3.6</td>
<td>3.0</td>
<td>2.7</td>
</tr>
<tr>
<td>15</td>
<td>3.2</td>
<td>3.0</td>
<td>6.8</td>
<td>5.8</td>
</tr>
<tr>
<td>16</td>
<td>2.2</td>
<td>2.8</td>
<td>3.0</td>
<td>4.6</td>
</tr>
<tr>
<td>17</td>
<td>2.8</td>
<td>2.7</td>
<td>4.6</td>
<td>4.5</td>
</tr>
<tr>
<td>18</td>
<td>3.1</td>
<td>2.8</td>
<td>6.5</td>
<td>7.2</td>
</tr>
<tr>
<td>19</td>
<td>2.7</td>
<td>3.5</td>
<td>5.0</td>
<td>2.8</td>
</tr>
<tr>
<td>20</td>
<td>2.4</td>
<td>3.0</td>
<td>4.0</td>
<td>4.6</td>
</tr>
<tr>
<td>21</td>
<td>2.6</td>
<td>2.5</td>
<td>5.0</td>
<td>4.3</td>
</tr>
<tr>
<td>22</td>
<td>2.7</td>
<td>2.6</td>
<td>5.0</td>
<td>4.0</td>
</tr>
<tr>
<td>23</td>
<td>3.1</td>
<td>2.9</td>
<td>5.1</td>
<td>2.1</td>
</tr>
<tr>
<td>24</td>
<td>2.3</td>
<td>2.8</td>
<td>7.0</td>
<td>6.0</td>
</tr>
<tr>
<td>25</td>
<td>2.6</td>
<td>2.9</td>
<td>6.0</td>
<td>5.4</td>
</tr>
<tr>
<td>26</td>
<td>2.8</td>
<td>3.3</td>
<td>4.5</td>
<td>6.3</td>
</tr>
<tr>
<td>27</td>
<td>2.9</td>
<td>2.4</td>
<td>3.6</td>
<td>2.6</td>
</tr>
<tr>
<td>28</td>
<td>3.2</td>
<td>2.5</td>
<td>2.8</td>
<td>7.8</td>
</tr>
<tr>
<td>29</td>
<td>2.7</td>
<td>2.3</td>
<td>4.5</td>
<td>6.1</td>
</tr>
<tr>
<td>30</td>
<td>2.6</td>
<td>2.7</td>
<td>7.0</td>
<td>6.7</td>
</tr>
<tr>
<td>31</td>
<td>3.4</td>
<td>2.5</td>
<td>6.1</td>
<td>5.3</td>
</tr>
<tr>
<td>32</td>
<td>3.2</td>
<td>3.0</td>
<td>3.9</td>
<td>2.8</td>
</tr>
<tr>
<td>33</td>
<td>2.1</td>
<td>2.6</td>
<td>5.6</td>
<td>3.6</td>
</tr>
<tr>
<td>34</td>
<td>2.7</td>
<td>2.8</td>
<td>6.5</td>
<td>6.5</td>
</tr>
<tr>
<td>35</td>
<td>2.4</td>
<td>2.5</td>
<td>4.5</td>
<td>5.0</td>
</tr>
<tr>
<td>36</td>
<td>3.0</td>
<td>2.5</td>
<td>3.8</td>
<td>4.5</td>
</tr>
<tr>
<td>37</td>
<td>2.3</td>
<td>3.0</td>
<td>4.5</td>
<td>3.1</td>
</tr>
<tr>
<td>38</td>
<td>2.8</td>
<td>2.6</td>
<td>3.2</td>
<td>3.2</td>
</tr>
<tr>
<td>39</td>
<td>2.1</td>
<td>2.3</td>
<td>4.8</td>
<td>5.4</td>
</tr>
<tr>
<td>40</td>
<td>2.3</td>
<td>3.3</td>
<td>3.4</td>
<td></td>
</tr>
<tr>
<td>41</td>
<td>2.9</td>
<td>3.3</td>
<td></td>
<td></td>
</tr>
<tr>
<td>42</td>
<td>2.5</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>43</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>44</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>45</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Mean: 2.75 2.88 4.67 4.58
### APPENDIX 1

(Continued)

#### Experiment 12

<table>
<thead>
<tr>
<th>Plant No.</th>
<th>Controls</th>
<th>Treatments</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>13-15</td>
<td>16-18</td>
</tr>
<tr>
<td>1</td>
<td>3.3</td>
<td>3.1</td>
</tr>
<tr>
<td>2</td>
<td>3.2</td>
<td>3.0</td>
</tr>
<tr>
<td>3</td>
<td>2.9</td>
<td>4.0</td>
</tr>
<tr>
<td>4</td>
<td>2.6</td>
<td>3.5</td>
</tr>
<tr>
<td>5</td>
<td>2.6</td>
<td>3.3</td>
</tr>
<tr>
<td>6</td>
<td>2.6</td>
<td>3.2</td>
</tr>
<tr>
<td>7</td>
<td>2.3</td>
<td>3.4</td>
</tr>
<tr>
<td>8</td>
<td>2.5</td>
<td>3.1</td>
</tr>
<tr>
<td>9</td>
<td>2.6</td>
<td>2.8</td>
</tr>
<tr>
<td>10</td>
<td>2.0</td>
<td>2.7</td>
</tr>
<tr>
<td>11</td>
<td>3.3</td>
<td>3.0</td>
</tr>
<tr>
<td>12</td>
<td>2.6</td>
<td>3.1</td>
</tr>
<tr>
<td>13</td>
<td>3.0</td>
<td>3.7</td>
</tr>
<tr>
<td>14</td>
<td>3.3</td>
<td>3.5</td>
</tr>
<tr>
<td>15</td>
<td>2.6</td>
<td>2.5</td>
</tr>
<tr>
<td>16</td>
<td>2.9</td>
<td>3.5</td>
</tr>
<tr>
<td>17</td>
<td>2.2</td>
<td>2.7</td>
</tr>
<tr>
<td>18</td>
<td>2.6</td>
<td>3.2</td>
</tr>
<tr>
<td>19</td>
<td>2.2</td>
<td>2.6</td>
</tr>
<tr>
<td>20</td>
<td>3.6</td>
<td>3.0</td>
</tr>
<tr>
<td>21</td>
<td>3.4</td>
<td>2.6</td>
</tr>
<tr>
<td>22</td>
<td>2.8</td>
<td>3.0</td>
</tr>
<tr>
<td>23</td>
<td>2.8</td>
<td>2.7</td>
</tr>
<tr>
<td>24</td>
<td>2.1</td>
<td>2.5</td>
</tr>
<tr>
<td>25</td>
<td>2.2</td>
<td>3.0</td>
</tr>
<tr>
<td>26</td>
<td>2.4</td>
<td>2.8</td>
</tr>
<tr>
<td>27</td>
<td>3.5</td>
<td>2.4</td>
</tr>
<tr>
<td>28</td>
<td>2.5</td>
<td>2.6</td>
</tr>
<tr>
<td>29</td>
<td>2.5</td>
<td>3.0</td>
</tr>
<tr>
<td>30</td>
<td>3.8</td>
<td>3.0</td>
</tr>
<tr>
<td>31</td>
<td>3.5</td>
<td>2.2</td>
</tr>
<tr>
<td>32</td>
<td>2.5</td>
<td>2.6</td>
</tr>
<tr>
<td>33</td>
<td>2.5</td>
<td>3.0</td>
</tr>
<tr>
<td>34</td>
<td>3.0</td>
<td>2.6</td>
</tr>
<tr>
<td>35</td>
<td>3.2</td>
<td>2.9</td>
</tr>
<tr>
<td>36</td>
<td>3.3</td>
<td>3.1</td>
</tr>
<tr>
<td>37</td>
<td>3.5</td>
<td>2.3</td>
</tr>
<tr>
<td>38</td>
<td>2.5</td>
<td>2.0</td>
</tr>
<tr>
<td>39</td>
<td>4.0</td>
<td>2.6</td>
</tr>
<tr>
<td>40</td>
<td>2.5</td>
<td>2.5</td>
</tr>
<tr>
<td>41</td>
<td>2.5</td>
<td>2.5</td>
</tr>
<tr>
<td>42</td>
<td>2.3</td>
<td>2.3</td>
</tr>
<tr>
<td>43</td>
<td>3.2</td>
<td>3.2</td>
</tr>
<tr>
<td>44</td>
<td></td>
<td></td>
</tr>
<tr>
<td>45</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Mean: 2.83 2.89 5.74 5.52
## Experiment 12

<table>
<thead>
<tr>
<th>Plant No.</th>
<th>Controls</th>
<th>Treatments</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Rolls 25-27</td>
<td>Rolls 28-30</td>
</tr>
<tr>
<td>1</td>
<td>2.5</td>
<td>2.5</td>
</tr>
<tr>
<td>2</td>
<td>2.2</td>
<td>2.8</td>
</tr>
<tr>
<td>3</td>
<td>2.9</td>
<td>3.4</td>
</tr>
<tr>
<td>4</td>
<td>3.0</td>
<td>2.2</td>
</tr>
<tr>
<td>5</td>
<td>2.4</td>
<td>3.8</td>
</tr>
<tr>
<td>6</td>
<td>2.9</td>
<td>2.6</td>
</tr>
<tr>
<td>7</td>
<td>2.8</td>
<td>3.2</td>
</tr>
<tr>
<td>8</td>
<td>3.5</td>
<td>2.9</td>
</tr>
<tr>
<td>9</td>
<td>3.0</td>
<td>2.9</td>
</tr>
<tr>
<td>10</td>
<td>3.0</td>
<td>3.3</td>
</tr>
<tr>
<td>11</td>
<td>3.2</td>
<td>3.2</td>
</tr>
<tr>
<td>12</td>
<td>3.0</td>
<td>3.6</td>
</tr>
<tr>
<td>13</td>
<td>2.9</td>
<td>3.1</td>
</tr>
<tr>
<td>14</td>
<td>4.0</td>
<td>2.9</td>
</tr>
<tr>
<td>15</td>
<td>2.8</td>
<td>2.9</td>
</tr>
<tr>
<td>16</td>
<td>3.4</td>
<td>3.1</td>
</tr>
<tr>
<td>17</td>
<td>2.8</td>
<td>3.4</td>
</tr>
<tr>
<td>18</td>
<td>2.8</td>
<td>2.7</td>
</tr>
<tr>
<td>19</td>
<td>2.6</td>
<td>2.4</td>
</tr>
<tr>
<td>20</td>
<td>2.5</td>
<td>3.1</td>
</tr>
<tr>
<td>21</td>
<td>3.6</td>
<td>2.7</td>
</tr>
<tr>
<td>22</td>
<td>3.4</td>
<td>2.6</td>
</tr>
<tr>
<td>23</td>
<td>2.2</td>
<td>3.1</td>
</tr>
<tr>
<td>24</td>
<td>3.6</td>
<td>3.8</td>
</tr>
<tr>
<td>25</td>
<td>2.2</td>
<td>2.3</td>
</tr>
<tr>
<td>26</td>
<td>3.4</td>
<td>3.1</td>
</tr>
<tr>
<td>27</td>
<td>2.2</td>
<td>2.8</td>
</tr>
<tr>
<td>28</td>
<td>2.2</td>
<td>3.6</td>
</tr>
<tr>
<td>29</td>
<td>3.2</td>
<td>3.2</td>
</tr>
<tr>
<td>30</td>
<td>2.8</td>
<td>3.0</td>
</tr>
<tr>
<td>31</td>
<td>2.9</td>
<td>3.8</td>
</tr>
<tr>
<td>32</td>
<td>2.6</td>
<td>2.6</td>
</tr>
<tr>
<td>33</td>
<td>3.0</td>
<td>2.1</td>
</tr>
<tr>
<td>34</td>
<td>3.4</td>
<td>3.4</td>
</tr>
<tr>
<td>35</td>
<td>2.8</td>
<td>3.1</td>
</tr>
<tr>
<td>36</td>
<td>3.6</td>
<td>2.9</td>
</tr>
<tr>
<td>37</td>
<td>2.7</td>
<td>2.8</td>
</tr>
<tr>
<td>38</td>
<td>3.1</td>
<td>2.9</td>
</tr>
<tr>
<td>39</td>
<td>2.1</td>
<td>2.5</td>
</tr>
<tr>
<td>40</td>
<td>2.9</td>
<td></td>
</tr>
<tr>
<td>41</td>
<td></td>
<td></td>
</tr>
<tr>
<td>42</td>
<td></td>
<td></td>
</tr>
<tr>
<td>43</td>
<td></td>
<td></td>
</tr>
<tr>
<td>44</td>
<td></td>
<td></td>
</tr>
<tr>
<td>45</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Mean: 

<table>
<thead>
<tr>
<th></th>
<th>Controls</th>
<th>Treatments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plant No.</td>
<td>2.90</td>
<td>2.99</td>
</tr>
<tr>
<td></td>
<td>5.96</td>
<td>5.89</td>
</tr>
</tbody>
</table>
### Washoe Experiment 13

<table>
<thead>
<tr>
<th>Plant No.</th>
<th>Controls</th>
<th>Treatments</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Rolls 1-3</td>
<td>Rolls 4-6</td>
</tr>
<tr>
<td>1</td>
<td>3.3</td>
<td>2.7</td>
</tr>
<tr>
<td>2</td>
<td>2.4</td>
<td>2.5</td>
</tr>
<tr>
<td>3</td>
<td>3.6</td>
<td>3.0</td>
</tr>
<tr>
<td>4</td>
<td>3.0</td>
<td>3.2</td>
</tr>
<tr>
<td>5</td>
<td>3.0</td>
<td>2.3</td>
</tr>
<tr>
<td>6</td>
<td>2.7</td>
<td>2.6</td>
</tr>
<tr>
<td>7</td>
<td>2.8</td>
<td>3.0</td>
</tr>
<tr>
<td>8</td>
<td>3.1</td>
<td>2.5</td>
</tr>
<tr>
<td>9</td>
<td>2.3</td>
<td>2.5</td>
</tr>
<tr>
<td>10</td>
<td>2.8</td>
<td>2.8</td>
</tr>
<tr>
<td>11</td>
<td>2.8</td>
<td>2.6</td>
</tr>
<tr>
<td>12</td>
<td>3.0</td>
<td>2.3</td>
</tr>
<tr>
<td>13</td>
<td>2.4</td>
<td>2.5</td>
</tr>
<tr>
<td>14</td>
<td>3.0</td>
<td>2.9</td>
</tr>
<tr>
<td>15</td>
<td>2.9</td>
<td>2.5</td>
</tr>
<tr>
<td>16</td>
<td>2.6</td>
<td>2.8</td>
</tr>
<tr>
<td>17</td>
<td>2.6</td>
<td>2.6</td>
</tr>
<tr>
<td>18</td>
<td>2.2</td>
<td>2.7</td>
</tr>
<tr>
<td>19</td>
<td>2.1</td>
<td>2.7</td>
</tr>
<tr>
<td>20</td>
<td>2.9</td>
<td>2.3</td>
</tr>
<tr>
<td>21</td>
<td>2.6</td>
<td>2.8</td>
</tr>
<tr>
<td>22</td>
<td>2.8</td>
<td>3.3</td>
</tr>
<tr>
<td>23</td>
<td>2.9</td>
<td>2.9</td>
</tr>
<tr>
<td>24</td>
<td>3.0</td>
<td>3.3</td>
</tr>
<tr>
<td>25</td>
<td>2.6</td>
<td>3.2</td>
</tr>
<tr>
<td>26</td>
<td>2.4</td>
<td>3.5</td>
</tr>
<tr>
<td>27</td>
<td>3.4</td>
<td>3.6</td>
</tr>
<tr>
<td>28</td>
<td>2.6</td>
<td>2.8</td>
</tr>
<tr>
<td>29</td>
<td>2.6</td>
<td>2.5</td>
</tr>
<tr>
<td>30</td>
<td>2.7</td>
<td>3.0</td>
</tr>
<tr>
<td>31</td>
<td>2.7</td>
<td>2.5</td>
</tr>
<tr>
<td>32</td>
<td>3.2</td>
<td>2.5</td>
</tr>
<tr>
<td>33</td>
<td>2.6</td>
<td>2.9</td>
</tr>
<tr>
<td>34</td>
<td>2.8</td>
<td>2.8</td>
</tr>
<tr>
<td>35</td>
<td>2.8</td>
<td>2.7</td>
</tr>
<tr>
<td>36</td>
<td>2.3</td>
<td>3.3</td>
</tr>
<tr>
<td>37</td>
<td>2.9</td>
<td>2.3</td>
</tr>
<tr>
<td>38</td>
<td>2.8</td>
<td>2.8</td>
</tr>
<tr>
<td>39</td>
<td>2.1</td>
<td>2.4</td>
</tr>
</tbody>
</table>

**Mean:**

<table>
<thead>
<tr>
<th>Rolls 1-3</th>
<th>Rolls 4-6</th>
<th>Rolls 7-9</th>
<th>Rolls 10-12</th>
<th>Rolls 13-15</th>
<th>Rolls 16-18</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.75</td>
<td>2.78</td>
<td>4.34</td>
<td>4.38</td>
<td>4.25</td>
<td>4.32</td>
</tr>
</tbody>
</table>
### APPENDIX 1

(Continued)

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2.7</td>
<td>3.1</td>
<td>6.8</td>
<td>5.0</td>
<td>6.8</td>
<td>2.6</td>
</tr>
<tr>
<td>2</td>
<td>2.3</td>
<td>2.9</td>
<td>3.1</td>
<td>4.8</td>
<td>2.8</td>
<td>5.0</td>
</tr>
<tr>
<td>3</td>
<td>2.5</td>
<td>2.3</td>
<td>3.8</td>
<td>5.7</td>
<td>3.8</td>
<td>5.3</td>
</tr>
<tr>
<td>4</td>
<td>3.2</td>
<td>2.5</td>
<td>7.2</td>
<td>4.8</td>
<td>3.3</td>
<td>7.6</td>
</tr>
<tr>
<td>5</td>
<td>2.8</td>
<td>2.8</td>
<td>3.5</td>
<td>6.8</td>
<td>4.5</td>
<td>4.8</td>
</tr>
<tr>
<td>6</td>
<td>2.6</td>
<td>2.9</td>
<td>2.8</td>
<td>4.5</td>
<td>7.0</td>
<td>6.8</td>
</tr>
<tr>
<td>7</td>
<td>2.6</td>
<td>2.7</td>
<td>6.4</td>
<td>5.4</td>
<td>4.8</td>
<td>5.7</td>
</tr>
<tr>
<td>8</td>
<td>3.0</td>
<td>3.4</td>
<td>7.8</td>
<td>7.7</td>
<td>6.3</td>
<td>6.3</td>
</tr>
<tr>
<td>9</td>
<td>3.0</td>
<td>2.9</td>
<td>5.0</td>
<td>5.8</td>
<td>7.2</td>
<td>4.0</td>
</tr>
<tr>
<td>10</td>
<td>2.7</td>
<td>2.6</td>
<td>7.0</td>
<td>4.8</td>
<td>4.0</td>
<td>6.8</td>
</tr>
<tr>
<td>11</td>
<td>2.7</td>
<td>2.6</td>
<td>5.4</td>
<td>5.5</td>
<td>3.5</td>
<td>4.5</td>
</tr>
<tr>
<td>12</td>
<td>2.2</td>
<td>2.5</td>
<td>6.0</td>
<td>6.3</td>
<td>4.0</td>
<td>4.8</td>
</tr>
<tr>
<td>13</td>
<td>3.1</td>
<td>2.2</td>
<td>5.3</td>
<td>3.2</td>
<td>6.0</td>
<td>6.8</td>
</tr>
<tr>
<td>14</td>
<td>2.9</td>
<td>2.6</td>
<td>4.0</td>
<td>3.8</td>
<td>5.8</td>
<td>2.6</td>
</tr>
<tr>
<td>15</td>
<td>3.1</td>
<td>2.3</td>
<td>4.0</td>
<td>2.4</td>
<td>4.0</td>
<td>6.4</td>
</tr>
<tr>
<td>16</td>
<td>3.3</td>
<td>2.5</td>
<td>2.8</td>
<td>5.3</td>
<td>4.0</td>
<td>4.5</td>
</tr>
<tr>
<td>17</td>
<td>2.6</td>
<td>2.7</td>
<td>4.5</td>
<td>2.6</td>
<td>5.3</td>
<td>2.5</td>
</tr>
<tr>
<td>18</td>
<td>2.3</td>
<td>3.1</td>
<td>7.0</td>
<td>6.3</td>
<td>6.0</td>
<td>5.6</td>
</tr>
<tr>
<td>19</td>
<td>3.1</td>
<td>3.0</td>
<td>4.0</td>
<td>4.5</td>
<td>5.4</td>
<td>5.4</td>
</tr>
<tr>
<td>20</td>
<td>3.1</td>
<td>3.3</td>
<td>4.0</td>
<td>2.6</td>
<td>5.5</td>
<td>2.4</td>
</tr>
<tr>
<td>21</td>
<td>3.4</td>
<td>2.5</td>
<td>6.0</td>
<td>2.5</td>
<td>5.4</td>
<td>2.9</td>
</tr>
<tr>
<td>22</td>
<td>2.3</td>
<td>2.3</td>
<td>6.5</td>
<td>2.4</td>
<td>7.0</td>
<td>6.4</td>
</tr>
<tr>
<td>23</td>
<td>3.0</td>
<td>2.6</td>
<td>5.5</td>
<td>2.9</td>
<td>4.0</td>
<td>7.7</td>
</tr>
<tr>
<td>24</td>
<td>3.2</td>
<td>2.5</td>
<td>4.8</td>
<td>4.8</td>
<td>6.8</td>
<td>2.9</td>
</tr>
<tr>
<td>25</td>
<td>2.8</td>
<td>3.1</td>
<td>5.4</td>
<td>3.0</td>
<td>3.8</td>
<td>5.8</td>
</tr>
<tr>
<td>26</td>
<td>3.0</td>
<td>2.5</td>
<td>5.5</td>
<td>4.8</td>
<td>3.5</td>
<td>4.8</td>
</tr>
<tr>
<td>27</td>
<td>2.8</td>
<td>2.2</td>
<td>3.8</td>
<td>5.0</td>
<td>5.5</td>
<td>2.4</td>
</tr>
<tr>
<td>28</td>
<td>2.5</td>
<td>2.7</td>
<td>3.3</td>
<td>5.0</td>
<td>7.8</td>
<td>4.8</td>
</tr>
<tr>
<td>29</td>
<td>2.5</td>
<td>3.2</td>
<td>4.8</td>
<td>7.0</td>
<td>6.4</td>
<td>5.5</td>
</tr>
<tr>
<td>30</td>
<td>2.8</td>
<td>2.2</td>
<td>6.3</td>
<td>4.6</td>
<td>6.5</td>
<td>4.4</td>
</tr>
<tr>
<td>31</td>
<td>2.9</td>
<td>3.0</td>
<td>3.8</td>
<td>7.6</td>
<td>3.0</td>
<td>4.6</td>
</tr>
<tr>
<td>32</td>
<td>2.6</td>
<td>2.9</td>
<td>5.8</td>
<td>6.8</td>
<td>5.8</td>
<td>6.3</td>
</tr>
<tr>
<td>33</td>
<td>2.5</td>
<td>2.8</td>
<td>3.0</td>
<td>4.0</td>
<td>6.0</td>
<td>5.0</td>
</tr>
<tr>
<td>34</td>
<td>2.6</td>
<td>3.0</td>
<td>3.5</td>
<td>6.8</td>
<td>2.8</td>
<td>5.1</td>
</tr>
<tr>
<td>35</td>
<td>2.7</td>
<td>3.0</td>
<td>3.8</td>
<td>6.4</td>
<td>3.5</td>
<td>3.2</td>
</tr>
<tr>
<td>36</td>
<td>2.7</td>
<td>2.8</td>
<td>5.6</td>
<td>4.0</td>
<td>4.0</td>
<td>5.0</td>
</tr>
<tr>
<td>37</td>
<td>2.6</td>
<td>2.8</td>
<td>2.9</td>
<td>3.8</td>
<td>3.8</td>
<td>4.0</td>
</tr>
<tr>
<td>38</td>
<td>3.0</td>
<td>2.6</td>
<td>6.4</td>
<td>6.3</td>
<td>6.3</td>
<td>6.8</td>
</tr>
<tr>
<td>39</td>
<td>3.2</td>
<td>2.3</td>
<td>2.4</td>
<td>4.0</td>
<td>4.0</td>
<td>4.0</td>
</tr>
<tr>
<td>40</td>
<td>2.5</td>
<td>2.4</td>
<td>3.0</td>
<td>7.2</td>
<td>3.0</td>
<td>7.2</td>
</tr>
<tr>
<td>41</td>
<td>2.5</td>
<td>2.7</td>
<td>4.6</td>
<td>5.1</td>
<td>5.1</td>
<td>4.6</td>
</tr>
<tr>
<td>42</td>
<td>2.5</td>
<td>2.4</td>
<td>4.0</td>
<td>4.0</td>
<td>4.0</td>
<td>4.0</td>
</tr>
<tr>
<td>43</td>
<td>2.2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>44</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>45</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

| Mean:     | 2.78                  | 2.69                  | 4.92                   | 4.82                   | 5.05                   | 4.95                   |
## APPENDIX 1

### Grimm

(Continued)

<table>
<thead>
<tr>
<th>Plant No.</th>
<th>Controls</th>
<th>Treatments</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>3.0</td>
<td>2.8</td>
</tr>
<tr>
<td>2</td>
<td>2.9</td>
<td>2.9</td>
</tr>
<tr>
<td>3</td>
<td>2.5</td>
<td>2.8</td>
</tr>
<tr>
<td>4</td>
<td>2.7</td>
<td>3.6</td>
</tr>
<tr>
<td>5</td>
<td>3.0</td>
<td>3.2</td>
</tr>
<tr>
<td>6</td>
<td>2.8</td>
<td>3.0</td>
</tr>
<tr>
<td>7</td>
<td>3.0</td>
<td>3.8</td>
</tr>
<tr>
<td>8</td>
<td>2.5</td>
<td>2.6</td>
</tr>
<tr>
<td>9</td>
<td>3.5</td>
<td>2.1</td>
</tr>
<tr>
<td>10</td>
<td>3.1</td>
<td>4.0</td>
</tr>
<tr>
<td>11</td>
<td>2.7</td>
<td>3.4</td>
</tr>
<tr>
<td>12</td>
<td>2.5</td>
<td>3.1</td>
</tr>
<tr>
<td>13</td>
<td>2.5</td>
<td>2.7</td>
</tr>
<tr>
<td>14</td>
<td>3.0</td>
<td>2.8</td>
</tr>
<tr>
<td>15</td>
<td>3.2</td>
<td>3.2</td>
</tr>
<tr>
<td>16</td>
<td>2.5</td>
<td>3.0</td>
</tr>
<tr>
<td>17</td>
<td>3.0</td>
<td>3.3</td>
</tr>
<tr>
<td>18</td>
<td>3.0</td>
<td>2.9</td>
</tr>
<tr>
<td>19</td>
<td>2.6</td>
<td>2.5</td>
</tr>
<tr>
<td>20</td>
<td>3.0</td>
<td>2.8</td>
</tr>
<tr>
<td>21</td>
<td>2.5</td>
<td>3.6</td>
</tr>
<tr>
<td>22</td>
<td>3.6</td>
<td>2.5</td>
</tr>
<tr>
<td>23</td>
<td>2.5</td>
<td>3.0</td>
</tr>
<tr>
<td>24</td>
<td>2.4</td>
<td>2.7</td>
</tr>
<tr>
<td>25</td>
<td>2.8</td>
<td>2.6</td>
</tr>
<tr>
<td>26</td>
<td>3.5</td>
<td>2.9</td>
</tr>
<tr>
<td>27</td>
<td>2.7</td>
<td>3.3</td>
</tr>
<tr>
<td>28</td>
<td>3.3</td>
<td>2.5</td>
</tr>
<tr>
<td>29</td>
<td>2.8</td>
<td>2.7</td>
</tr>
<tr>
<td>30</td>
<td>3.6</td>
<td>2.8</td>
</tr>
<tr>
<td>31</td>
<td>3.3</td>
<td>2.6</td>
</tr>
<tr>
<td>32</td>
<td>2.9</td>
<td>3.5</td>
</tr>
<tr>
<td>33</td>
<td>2.6</td>
<td>2.9</td>
</tr>
<tr>
<td>34</td>
<td>2.7</td>
<td>2.4</td>
</tr>
<tr>
<td>35</td>
<td>2.9</td>
<td>2.8</td>
</tr>
<tr>
<td>36</td>
<td>3.3</td>
<td>2.7</td>
</tr>
<tr>
<td>37</td>
<td>3.8</td>
<td>3.1</td>
</tr>
<tr>
<td>38</td>
<td>2.3</td>
<td>3.3</td>
</tr>
<tr>
<td>39</td>
<td>2.9</td>
<td>2.2</td>
</tr>
<tr>
<td>40</td>
<td>2.8</td>
<td>2.8</td>
</tr>
<tr>
<td>41</td>
<td></td>
<td></td>
</tr>
<tr>
<td>42</td>
<td></td>
<td></td>
</tr>
<tr>
<td>43</td>
<td></td>
<td></td>
</tr>
<tr>
<td>44</td>
<td></td>
<td></td>
</tr>
<tr>
<td>45</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Mean: 2.91 2.94 5.41 5.27 5.37 5.45
## APPENDIX 2

### STEM DIAMETERS OF INOCULATED LUCERNE SEEDLINGS

**Tested for Nematode Reproduction in mm.**

<table>
<thead>
<tr>
<th>Reproduction Occurring</th>
<th>Normal</th>
<th>Nematodes Only</th>
</tr>
</thead>
<tbody>
<tr>
<td>6.0</td>
<td>2.7</td>
<td>3.4</td>
</tr>
<tr>
<td>7.5</td>
<td>3.8</td>
<td>4.2</td>
</tr>
<tr>
<td>3.5</td>
<td>3.2</td>
<td>4.0</td>
</tr>
<tr>
<td>4.5</td>
<td>2.1</td>
<td>3.5</td>
</tr>
<tr>
<td>5.2</td>
<td>2.6</td>
<td>3.5</td>
</tr>
<tr>
<td>5.4</td>
<td>2.7</td>
<td>3.0</td>
</tr>
<tr>
<td>2.6</td>
<td>2.3</td>
<td>2.3</td>
</tr>
<tr>
<td></td>
<td>2.2</td>
<td>5.0</td>
</tr>
<tr>
<td></td>
<td>2.3</td>
<td>4.5</td>
</tr>
<tr>
<td>Total number of plants 7</td>
<td>3.5</td>
<td>3.5</td>
</tr>
<tr>
<td></td>
<td>3.3</td>
<td>5.5</td>
</tr>
<tr>
<td></td>
<td>2.1</td>
<td>3.5</td>
</tr>
<tr>
<td></td>
<td>2.9</td>
<td>4.5</td>
</tr>
<tr>
<td>Mean stem diameter 4.96</td>
<td>3.4</td>
<td>5.4</td>
</tr>
<tr>
<td></td>
<td>3.5</td>
<td>4.3</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Normal</th>
<th>Nematodes Only</th>
</tr>
</thead>
<tbody>
<tr>
<td>4.0</td>
<td>5.0</td>
</tr>
<tr>
<td>3.5</td>
<td>3.5</td>
</tr>
<tr>
<td>4.5</td>
<td>4.0</td>
</tr>
<tr>
<td>3.8</td>
<td>3.8</td>
</tr>
<tr>
<td>4.5</td>
<td>4.5</td>
</tr>
<tr>
<td>3.4</td>
<td>3.4</td>
</tr>
<tr>
<td>3.5</td>
<td>3.5</td>
</tr>
<tr>
<td>3.0</td>
<td>3.0</td>
</tr>
<tr>
<td>Total number of plants 15</td>
<td>2.5</td>
</tr>
<tr>
<td></td>
<td>3.5</td>
</tr>
<tr>
<td></td>
<td>6.0</td>
</tr>
<tr>
<td>Mean stem diameter 2.84</td>
<td>5.5</td>
</tr>
<tr>
<td></td>
<td>6.0</td>
</tr>
<tr>
<td></td>
<td>3.2</td>
</tr>
<tr>
<td></td>
<td>4.3</td>
</tr>
<tr>
<td></td>
<td>4.0</td>
</tr>
<tr>
<td></td>
<td>3.0</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Normal</th>
<th>Nematodes Only</th>
</tr>
</thead>
<tbody>
<tr>
<td>3.0</td>
<td>3.0</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Total number of plants 51</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean stem diameter 3.92</td>
</tr>
</tbody>
</table>
## APPENDIX 2

(Continued)

<table>
<thead>
<tr>
<th>Wairau</th>
<th>Reproduction Occurring</th>
<th>Normal</th>
<th>Nematodes Only</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5.0</td>
<td>6.5 5.8</td>
<td>2.0</td>
<td>3.8</td>
</tr>
<tr>
<td>6.5</td>
<td>6.0 5.2</td>
<td>3.0</td>
<td>3.2</td>
</tr>
<tr>
<td>5.5</td>
<td>4.5 4.9</td>
<td>4.6</td>
<td>5.0</td>
</tr>
<tr>
<td>5.0</td>
<td>4.3 5.1</td>
<td>3.2</td>
<td>4.3</td>
</tr>
<tr>
<td>4.5</td>
<td>5.0 4.9</td>
<td>2.6</td>
<td>2.2</td>
</tr>
<tr>
<td>5.5</td>
<td>4.5 5.8</td>
<td>2.2</td>
<td>2.4</td>
</tr>
<tr>
<td>5.5</td>
<td>7.0 4.1</td>
<td>3.7</td>
<td>3.5</td>
</tr>
<tr>
<td>7.0</td>
<td>4.5 7.8</td>
<td>3.0</td>
<td>3.0</td>
</tr>
<tr>
<td>5.9</td>
<td>6.2 4.6</td>
<td>3.5</td>
<td>3.5</td>
</tr>
<tr>
<td>3.0</td>
<td>5.0 5.2</td>
<td>4.0</td>
<td>4.8</td>
</tr>
<tr>
<td>6.0</td>
<td>4.3 4.0</td>
<td>3.0</td>
<td>5.0</td>
</tr>
<tr>
<td>6.0</td>
<td>6.0 3.4</td>
<td>3.1</td>
<td>3.9</td>
</tr>
<tr>
<td>7.0</td>
<td>6.5 3.1</td>
<td>2.8</td>
<td>6.5</td>
</tr>
<tr>
<td>6.0</td>
<td>5.6 6.1</td>
<td>4.8</td>
<td>5.5</td>
</tr>
<tr>
<td>5.5</td>
<td>4.6 2.5</td>
<td>2.5</td>
<td>2.8</td>
</tr>
<tr>
<td>6.0</td>
<td>4.0 4.0</td>
<td>4.0</td>
<td>5.5</td>
</tr>
<tr>
<td>7.0</td>
<td>5.0 2.9</td>
<td>2.6</td>
<td>5.0</td>
</tr>
<tr>
<td>4.5</td>
<td>5.2 2.6</td>
<td>3.2</td>
<td>2.7</td>
</tr>
<tr>
<td>6.5</td>
<td>4.5 3.2</td>
<td>3.7</td>
<td>4.1</td>
</tr>
<tr>
<td>5.5</td>
<td>5.0 2.8</td>
<td>2.8</td>
<td>4.5</td>
</tr>
<tr>
<td>5.0</td>
<td>4.2 2.7</td>
<td>4.0</td>
<td>4.5</td>
</tr>
<tr>
<td>6.5</td>
<td>6.2 4.0</td>
<td>3.3</td>
<td>2.5</td>
</tr>
<tr>
<td>5.5</td>
<td>4.2 3.3</td>
<td>4.5</td>
<td>4.3</td>
</tr>
<tr>
<td>4.5</td>
<td>7.0 2.5</td>
<td>4.5</td>
<td>2.5</td>
</tr>
<tr>
<td>6.5</td>
<td>4.9 4.5</td>
<td>4.5</td>
<td>4.8</td>
</tr>
<tr>
<td>6.3</td>
<td>6.3 2.4</td>
<td>4.0</td>
<td>3.0</td>
</tr>
<tr>
<td>6.0</td>
<td>4.7 4.6</td>
<td>4.0</td>
<td>4.6</td>
</tr>
<tr>
<td>6.5</td>
<td>3.2 5.3</td>
<td>2.0</td>
<td>5.3</td>
</tr>
<tr>
<td>4.5</td>
<td>5.6 4.3</td>
<td>2.0</td>
<td>3.7</td>
</tr>
</tbody>
</table>

Total number of plants 70

Total number of plants 29

Total number of plants 32

Mean stem diameter 5.43

Mean stem diameter 3.22

Mean stem diameter 4.03
APPENDIX 2

(Continued)

<table>
<thead>
<tr>
<th>Grimm Reproduction Occurring</th>
<th>Normal</th>
<th>Nematodes Only</th>
</tr>
</thead>
<tbody>
<tr>
<td>7.3 7.0</td>
<td>5.5</td>
<td>4.8</td>
</tr>
<tr>
<td>10.0 8.5</td>
<td>3.0</td>
<td>4.8</td>
</tr>
<tr>
<td>5.8 7.0</td>
<td>2.8</td>
<td>5.0</td>
</tr>
<tr>
<td>5.5 7.6</td>
<td>5.0</td>
<td>4.0</td>
</tr>
<tr>
<td>6.0 6.7</td>
<td>3.0</td>
<td>4.7</td>
</tr>
<tr>
<td>4.5 6.7</td>
<td>4.3</td>
<td>6.5</td>
</tr>
<tr>
<td>4.0 4.8</td>
<td>4.3</td>
<td>5.0</td>
</tr>
<tr>
<td>7.0 5.8</td>
<td>2.3</td>
<td>4.5</td>
</tr>
<tr>
<td>8.0 8.0</td>
<td>2.3</td>
<td>7.5</td>
</tr>
<tr>
<td>6.0 5.1</td>
<td>3.2</td>
<td>5.0</td>
</tr>
<tr>
<td>5.3 6.8</td>
<td>2.5</td>
<td>5.5</td>
</tr>
<tr>
<td>5.7 6.9</td>
<td>4.0</td>
<td>4.0</td>
</tr>
<tr>
<td>6.5 7.5</td>
<td>4.0</td>
<td>6.0</td>
</tr>
<tr>
<td>6.0 6.0</td>
<td>4.0</td>
<td>6.0</td>
</tr>
<tr>
<td>7.0 6.8</td>
<td>2.0</td>
<td>4.0</td>
</tr>
<tr>
<td>6.5 5.0</td>
<td>2.6</td>
<td>3.1</td>
</tr>
<tr>
<td>6.5</td>
<td>3.8</td>
<td>4.6</td>
</tr>
<tr>
<td>5.0</td>
<td>3.5</td>
<td></td>
</tr>
<tr>
<td>6.0</td>
<td>3.8</td>
<td></td>
</tr>
<tr>
<td>7.0</td>
<td>3.5</td>
<td></td>
</tr>
<tr>
<td>6.0</td>
<td>2.9</td>
<td></td>
</tr>
<tr>
<td>5.8</td>
<td>2.2</td>
<td></td>
</tr>
<tr>
<td>6.5</td>
<td>2.8</td>
<td></td>
</tr>
<tr>
<td>6.5</td>
<td>3.4</td>
<td></td>
</tr>
<tr>
<td>8.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>4.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6.0</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Total number of plants 17

Mean stem diameter 5.00

Total number of plants 24

Mean stem diameter 3.36

Total number of plants 46

Mean stem diameter 6.43
APPENDIX 2

(Continued)

<table>
<thead>
<tr>
<th>Washoe</th>
<th>Experiment 12</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reproduction Occurring</td>
<td>Normal</td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>4.0</td>
<td>3.1</td>
</tr>
<tr>
<td>7.8</td>
<td>3.0</td>
</tr>
<tr>
<td>5.6</td>
<td>3.6</td>
</tr>
<tr>
<td>5.3</td>
<td>2.8</td>
</tr>
<tr>
<td>4.1</td>
<td>3.2</td>
</tr>
<tr>
<td>6.0</td>
<td>3.4</td>
</tr>
<tr>
<td>3.5</td>
<td>2.5</td>
</tr>
<tr>
<td>6.8</td>
<td>4.5</td>
</tr>
<tr>
<td>6.5</td>
<td>2.7</td>
</tr>
<tr>
<td>5.0</td>
<td>2.8</td>
</tr>
<tr>
<td>4.0</td>
<td>2.7</td>
</tr>
<tr>
<td>5.0</td>
<td>2.8</td>
</tr>
<tr>
<td>5.0</td>
<td>2.1</td>
</tr>
<tr>
<td>5.1</td>
<td>2.6</td>
</tr>
<tr>
<td>7.0</td>
<td>2.8</td>
</tr>
<tr>
<td>7.0</td>
<td>3.7</td>
</tr>
<tr>
<td>6.1</td>
<td></td>
</tr>
<tr>
<td>5.6</td>
<td></td>
</tr>
<tr>
<td>6.5</td>
<td></td>
</tr>
<tr>
<td>4.5</td>
<td></td>
</tr>
<tr>
<td>7.3</td>
<td></td>
</tr>
<tr>
<td>6.0</td>
<td></td>
</tr>
<tr>
<td>6.5</td>
<td></td>
</tr>
<tr>
<td>5.8</td>
<td></td>
</tr>
<tr>
<td>7.2</td>
<td></td>
</tr>
<tr>
<td>6.0</td>
<td></td>
</tr>
<tr>
<td>5.4</td>
<td></td>
</tr>
<tr>
<td>6.3</td>
<td></td>
</tr>
<tr>
<td>7.8</td>
<td></td>
</tr>
<tr>
<td>6.1</td>
<td></td>
</tr>
<tr>
<td>6.7</td>
<td></td>
</tr>
<tr>
<td>4.5</td>
<td></td>
</tr>
</tbody>
</table>

Total number of plants 16
Mean stem diameter 3.19

Total number of plants 32
Mean stem diameter 5.81

Total number of plants 29
Mean stem diameter 4.25
### APPENDIX 2

*(Continued)*

<table>
<thead>
<tr>
<th>Reproduction Occurring</th>
<th>Normal</th>
<th>Nematodes Only</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8.0 7.2</td>
<td>3.5</td>
<td>5.0</td>
</tr>
<tr>
<td>8.0 6.9</td>
<td>4.0</td>
<td>4.5</td>
</tr>
<tr>
<td>7.6 7.2</td>
<td>2.4</td>
<td>2.4</td>
</tr>
<tr>
<td>5.0 5.2</td>
<td>2.4</td>
<td>2.4</td>
</tr>
<tr>
<td>7.4 5.6</td>
<td>3.6</td>
<td>4.5</td>
</tr>
<tr>
<td>5.5 6.5</td>
<td>2.8</td>
<td>5.0</td>
</tr>
<tr>
<td>6.5 6.3</td>
<td>2.4</td>
<td>4.0</td>
</tr>
<tr>
<td>6.0 7.2</td>
<td>3.5</td>
<td>5.0</td>
</tr>
<tr>
<td>6.8 4.7</td>
<td>3.2</td>
<td>6.5</td>
</tr>
<tr>
<td>7.0 7.8</td>
<td></td>
<td>4.0</td>
</tr>
<tr>
<td>7.2 6.8</td>
<td></td>
<td>4.7</td>
</tr>
<tr>
<td>7.5 6.6</td>
<td></td>
<td>4.0</td>
</tr>
<tr>
<td>6.3 6.7</td>
<td></td>
<td>5.2</td>
</tr>
<tr>
<td>6.8 8.0</td>
<td></td>
<td>5.8</td>
</tr>
<tr>
<td>6.0 6.4</td>
<td></td>
<td>6.0</td>
</tr>
<tr>
<td>6.3 6.0</td>
<td></td>
<td>5.8</td>
</tr>
<tr>
<td>5.5 4.8</td>
<td></td>
<td>4.0</td>
</tr>
<tr>
<td>8.0</td>
<td></td>
<td>5.2</td>
</tr>
<tr>
<td>6.3</td>
<td></td>
<td>4.8</td>
</tr>
<tr>
<td>6.5</td>
<td></td>
<td>3.7</td>
</tr>
<tr>
<td>6.5</td>
<td></td>
<td>4.0</td>
</tr>
<tr>
<td>6.0</td>
<td></td>
<td>5.3</td>
</tr>
<tr>
<td>6.7</td>
<td></td>
<td>5.9</td>
</tr>
<tr>
<td>8.0</td>
<td></td>
<td>4.0</td>
</tr>
<tr>
<td>7.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5.6</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7.2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>4.6</td>
<td></td>
<td></td>
</tr>
<tr>
<td>4.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5.6</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Total number of plants 24

Mean stem diameter 4.82

Total number of plants 47

Mean stem diameter 6.52
<table>
<thead>
<tr>
<th>Grimm</th>
<th>Normal</th>
<th>Nematodes Only</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reproduction Occurring</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6.0 6.5</td>
<td>3.5</td>
<td>4.0</td>
</tr>
<tr>
<td>8.0 8.3</td>
<td>3.0</td>
<td>3.0</td>
</tr>
<tr>
<td>7.5 6.5</td>
<td>6.0</td>
<td>5.7</td>
</tr>
<tr>
<td>9.0 7.2</td>
<td>2.5</td>
<td>2.9</td>
</tr>
<tr>
<td>6.0 6.4</td>
<td>2.5</td>
<td>3.6</td>
</tr>
<tr>
<td>6.0 7.2</td>
<td>3.6</td>
<td>5.5</td>
</tr>
<tr>
<td>6.6 7.5</td>
<td>2.3</td>
<td>6.0</td>
</tr>
<tr>
<td>9.6 6.0</td>
<td>3.6</td>
<td>4.0</td>
</tr>
<tr>
<td>8.3 7.0</td>
<td></td>
<td>5.6</td>
</tr>
<tr>
<td>6.3 8.0</td>
<td></td>
<td>4.5</td>
</tr>
<tr>
<td>6.5 6.5</td>
<td></td>
<td>5.0</td>
</tr>
<tr>
<td>6.8 5.5</td>
<td></td>
<td>5.8</td>
</tr>
<tr>
<td>7.0 6.5</td>
<td></td>
<td>5.0</td>
</tr>
<tr>
<td>6.0 8.6</td>
<td></td>
<td>5.2</td>
</tr>
<tr>
<td>4.0 7.5</td>
<td>Total number of plants 8</td>
<td>Total number of plants 21</td>
</tr>
<tr>
<td>6.0 7.3</td>
<td>Mean stem diameter 3.38</td>
<td>Mean stem diameter 4.74</td>
</tr>
<tr>
<td>6.0 6.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7.0 5.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>8.3 6.2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5.6 6.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5.5 7.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6.5 6.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>8.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6.3</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total number of plants 52</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean stem diameter 6.79</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### APPENDIX 2

(Continued)

<table>
<thead>
<tr>
<th>Washoe</th>
<th>Experiment 13</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Reproduction Occurring</td>
</tr>
<tr>
<td></td>
<td>5.8</td>
</tr>
<tr>
<td></td>
<td>5.2</td>
</tr>
<tr>
<td></td>
<td>4.2</td>
</tr>
<tr>
<td></td>
<td>5.2</td>
</tr>
<tr>
<td></td>
<td>5.6</td>
</tr>
<tr>
<td></td>
<td>5.0</td>
</tr>
<tr>
<td></td>
<td>5.2</td>
</tr>
<tr>
<td></td>
<td>3.3</td>
</tr>
<tr>
<td></td>
<td>3.6</td>
</tr>
<tr>
<td></td>
<td>6.2</td>
</tr>
<tr>
<td></td>
<td>5.8</td>
</tr>
<tr>
<td></td>
<td>5.2</td>
</tr>
<tr>
<td></td>
<td>7.3</td>
</tr>
<tr>
<td></td>
<td>6.4</td>
</tr>
<tr>
<td></td>
<td>5.2</td>
</tr>
<tr>
<td></td>
<td>4.5</td>
</tr>
<tr>
<td></td>
<td>5.6</td>
</tr>
<tr>
<td></td>
<td>4.2</td>
</tr>
<tr>
<td></td>
<td>6.3</td>
</tr>
<tr>
<td></td>
<td>5.0</td>
</tr>
<tr>
<td></td>
<td>5.8</td>
</tr>
<tr>
<td></td>
<td>4.6</td>
</tr>
<tr>
<td></td>
<td>5.0</td>
</tr>
<tr>
<td></td>
<td>4.2</td>
</tr>
<tr>
<td></td>
<td>4.5</td>
</tr>
<tr>
<td></td>
<td>6.3</td>
</tr>
<tr>
<td></td>
<td>5.8</td>
</tr>
<tr>
<td></td>
<td>5.4</td>
</tr>
</tbody>
</table>

Total number of plants 28

Mean stem diameter 5.23

Total number of plants 26

Mean stem diameter 4.19
## APPENDIX 2

(Continued)

### Wairau

<table>
<thead>
<tr>
<th>Reproduction Occurring</th>
<th>Normal</th>
<th>Nematodes only</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>3.1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>3.5</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.8</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.8</td>
</tr>
<tr>
<td></td>
<td></td>
<td>3.5</td>
</tr>
<tr>
<td></td>
<td></td>
<td>3.2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>3.8</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.4</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.6</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.6</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.5</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.4</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.0</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.9</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.4</td>
</tr>
<tr>
<td>Total number of plants</td>
<td>15</td>
<td></td>
</tr>
<tr>
<td>Mean stem diameter</td>
<td>2.97</td>
<td></td>
</tr>
</tbody>
</table>

### Experiment 13

|                         |       | 4.6            | 4.0 |
| Total number of plants | 19    |                | 19  |
| Mean stem diameter     | 3.95  |                | 3.95|

Total number of plants 45

Mean stem diameter 5.84