The Biology of Proterodiplosis radicis Wyatt
(Cecidomyiidae : Diptera)

A Thesis presented in partial fulfilment
of the requirements for the degree of
M.Sc in Zoology at
Massey University

MALCOLM KNOX KAY
1974

ABSTRACT

Aspects of the biology of Proterodiplosis radicis were investigated. Field studies in the S.W. Ruahine Range were supplemented with laboratory experimentation.

P.radicis is a root parasite of Metrosideros robusta A. Cunn. (MYRTACEAE), and forms galls on the tree's fine rootlets. The host's phenology and the developmental stages of P.radicis are described. The insect passes through three distinct larval instars and pupates within the gall. Adult emergence continues from mid-February to March. The adults represent a non-feeding, short-lived (2-4 days) reproductive stage in the life cycle. Dispersal appears to be heterogenous and of little direct consequence to the population. Other aspects of the dynamics of the population were investigated and the findings collated in a life table. Greatest mortality occurs while the insect is without the protection of the gall, i.e. the adults, eggs and first larval instar. The first larval instar is of long duration and predation by eurytopic soil fauna probably effects the most significant regulation of the insect's numbers. The pupa is parasitised by a small ceraphronid while within the gall.

P.radicis was found to be strongly host-specific. The subtleties of the intimate plant-insect association are discussed with reference to gall structure, metabolism and chemistry. The conclusion is drawn that P.radicis cannot be seriously implicated in the widespread debility of Metrosideros robusta.

PREFACE

The thesis investigates various aspects of the biology of *Proterodiplosis radicis* - a root parasite of *Metrosideros robusta*. It is presented in three parts.

The first part describes the area in which the study was undertaken, the host tree, the phenology of the host and the developmental stages of *P.radicis*. The second part describes some aspects of the biology of *P.radicis* and factors influencing the dynamics of the insect's population. This section culminates in a life-table where the impact of the insect's biological character and external negating factors are assessed.

The third part attempts to describe the finer points of an intimate insect-plant association. The significance of the interaction to the biologies of the host and parasite is discussed.

Acknowledgements

I wish to thank Dr R.H. Milnes for his advice and encouragement in supervising this work and for his helpful criticism of the manuscript. I also thank the staff of the Botany-Zoology Department, Massey University, for their stimulating discussions. Advice on computer and statistical matters was freely given by Dr C. Boswell and Mr Arnold of the Maths Department.

Thanks are also due to my wife who assisted with tedious field work and to Mrs F. Norrie for her diligence in typing the manuscript.

I am indebted to the New Zealand Forest Service for providing a grant to cover equipment and transport costs.

CONTENTS

PAGE

PREFACE AND ACKNOWLEDGEMENTS iii

PART I A DESCRIPTION OF THE HOST AND THE PARASITE

1 Introduction 1

2 The Study Area (Description and Meterology) 4

3 The Host, Northern Rata (*Metrosideros robusta* A. Cunn. MYRTACEAE) 7

4 The Description of *Proterodiplosis radicis* Wyatt 18

PART II THE BIOLOGY OF *PROTERODIPLOSIS RADICIS*

1 Introduction 23

2 The Sampling Programme 26

3 The Life Cycle of *P.radicis* 32

4 Adult Longevity, Mating and Fecundity 37

5 Distribution and dispersion of *P.radicis* 44

6 Dispersal 54

7 The Influence of Weather on the Population Dynamics of *P.radicis* 58

8 Intra-specific Competition 63

9 Mortality - predation of *P.radicis* 66

10 Mortality - parasitism of *P.radicis* 73

11 A Life Table for *P.radicis* 78

PART III THE INSECT-PLANT RELATIONSHIP

1 Introduction 81

2 Host Specificity 86

3 Gall Structure 91

4 Gall Metabolism 95

5 Gall Chemistry - the Host's Defence 98

PAGE

GENERAL CONCLUSION 102

SUMMARY 111

APPENDICES 113

BIBLIOGRAPHY 141

LIST OF FIGURES AND PLATES

FIGURE NO.		AFTER PAGE
1	Map of Ruahine Range	2
2	Map of study area	6
3	Meteorological record	6
4	Typical thermohydrograph record	6
5	Rata phenology	12
6	Rata mortality	16
7	Head capsule histogram	20
8	The larvae of <u>P.radicis</u>	21
9	<u>P.radicis</u> adult male	22
10	Antennal segments and genitalia of adult <u>P.radicis</u>	22
11	Sampling grid	28
12	Phenolic chromatographs	30
13	Population structure	33
14	Larva-gall regression	33
15	Gall size distribution	33
16	Sternal spatula tine length	35
17	North Island distribution of <u>P.radicis</u>	49
18	Root distribution	50
19	Gall distribution	50
20	Root density with respect to soil depth	50
21	Gall " " " " " "	50
22	Root and gall dispersion	50
23	Desiccation rates	61
24	Adult female ceraphronid	74
25	Juvenile stages of ceraphronid	75

NO.		AFTER PAGE
26	Mouthparts of nematode predator	72
27	The relationship between pupal and parasite density	77
28	Survivorship curve	79
29	Root sections of <u>Metrosideros</u> species	89
30	Host specificity	89
31	Comparative gall structure	93
32	Respiration rates	97

PLATE NO.

1	The study area	6
2	Galls on fine roots of <u>M.robusta</u>	9
3	'Runner' roots of <u>M.robusta</u>	9
4	Growth form of <u>M.robusta</u>	9
5	An increment band	12
6	<u>M.perforata</u>	27
7	<u>M.diffusa</u>	27
8	Multiple refluxer	29
9	Rata mortality	15
10	Malaise trap	30
11	Sex attraction trap	39
12a,b	Control and baited trap	39
13	High gall density	63
14	Predatory nematode	72
15	Transverse section of gall	92

INTRODUCTION

The Cecidomyiidae represent a large family of nematocerous Diptera. Although small in size the 5000 or so recorded species, with their novel habits, make this a conspicuous element in the world's insect fauna. They are most closely related to the fungivorous Mycetophilidae and a number of the more generalised cecids share this habit. Certain other cecids, notably Aphidoletes and Mycodiptermis, are important as predators of other insects, especially Homoptera. However, the greater majority are phytophagous and may cause conspicuous, localised plant deformities, or galls, during their association with the plant. Many have become serious economic pests with some attaining international pest status. The Hessian fly (Meyetiola destructor), pear midge (Contarinia pyrivora) and clover midge (Dasineura leguminicola) are common pests throughout the world. Adult cecids are small two-winged flies ranging in size from 0.5-8.0 mm (Felt, 1925). They are readily dispersed (Johnson, 1969) and the phytophagous larvae are so well protected that control is often difficult.

The Cecidomyiidae, or gall midges, of economic importance in New Zealand have all been introduced. Our knowledge of endemic species is fragmentary. The Southern Hemisphere gall midge fauna appears to contain a considerable number of ancient, highly specialised, offshoots from the more primitive cosmopolitan genera, such as Lastremia, Porricendyla and Dasineura (Felt, 1925). Their character suggests a long period of zoological isolation. Lamb (1962) and Barnes (1937) have recorded a total of 40 species from New Zealand. The majority of these were captured as adults and tentatively identified with little knowledge of their biologies. Only the more common and economically significant species have so far been studied in detail.

Proterodiplosis radicis Wyatt was discovered during an investigation into the widespread mortality of northern rata (Metrosideros robusta A. Cunn.), a dominant indigenous forest species.

The midge proved not only to be a new species, but also a new genus of some economic interest (Wyatt, 1963). On the character of the male antennal structure, the midge was considered to be a primitive member of the tribe *Cecidomyiidi trifila*, other members of which show a range of feeding habits from predation to phytophagy. *P.radicis*, however, has the comparatively rare habit (Mani, 1964) of forming galls on the fine rootlets of the tree. The galls were found to occur on the roots of northern rata over a large area of the central North Island. However, it was considered improbable that they were an important factor in the mortality of the tree (Wyatt, 1963).

Northern rata is distributed throughout the North Island and the north of the South Island. Typically the seedling tree is an epiphyte, usually of rimu (*Dacrydium cupressinum*). Its descending ground roots coalesce and gradually overgrow the host tree. Over a number of years the host decays, leaving a massive, irregular, and to some extent, hollow tree. Its irregular epiphytic growth forms create conversion and seasoning difficulties for timber millers so the tree has no substantial market other than for local consumption as firewood. For these reasons rata is rarely felled and has become of some importance in watershed maintenance in milled lower montane forests (Masters *et al.*, 1957).

This study was conducted in the Southern Ruahine Range. As early as 1951 heavy rata mortality was recorded in the Tamaki Valley (S.E. Ruahines, Fig. 1) and by 1952 canopy defoliation was conspicuous from the plains (Elder, 1958). Early in 1955 rata and kamahi (*Weinmannia racemosa*) mortality was reported from several areas almost simultaneously and an interdepartmental survey was instigated (Holloway *et al.*, 1955). The survey revealed a number of injurious agents affecting rata. Individually these factors were capable of inflicting localised mortality; however, the widespread damage encountered was considered to be a complex ecological problem (Hoy *et al.*, 1955). The present study investigates the role of *P.radicis* within this complex.

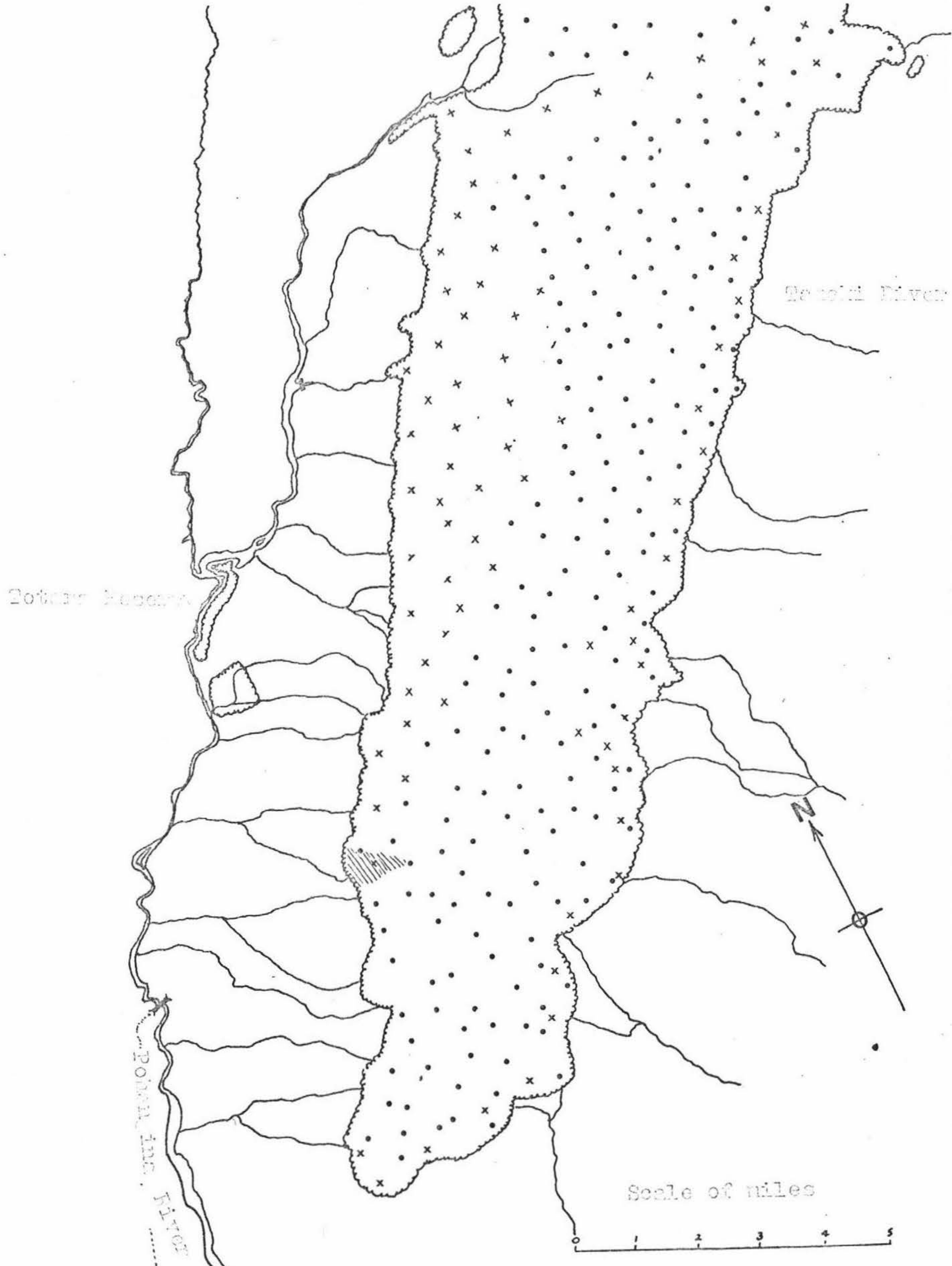


Fig.1 The Ruahine Range; ▨ indicates the position of the stu
area. (after Elder 1965)

The study of gall midge biology is not easy. Owing to their small size and fragility they are difficult to detect and handle in the field and often there is a close resemblance between different genera (Felt, 1925). The absence of any data on P.radicis other than a taxonomic description, and the insect's intimate involvement with a host equally lacking in phenological description, left a wide field in which it was desirable to gain as much knowledge as possible in order to make valid conclusions about the effect of the parasite. Using a sequential sampling programme, it was possible to elucidate, simultaneously, the insect's life-cycle, various regulating factors, and some aspects of rata phenology. The life-cycle and ecological factors affecting it were summed in a life-table, and the field work was complemented by laboratory studies to assess the significance of the insect-plant association. The study was initiated in 1971 but the bulk of the field work took place between January 1972 and March 1973.

The study was acknowledged and assisted by a research grant from the New Zealand Forest Research Institute.

The Study Area - (Description and Meteorology)

The study area (lat.40°12', long.175°53'), situated some 25 miles north of Palmerston North on the south-west of the Ruahine Ranges, was chosen for convenience rather than any biological or statistical attributes. The area possessed good access to a relatively undisturbed area of forest with a reasonable number of northern rata (Fig.2).

The Ruahine Range is a portion of the main North Island divide and extends 56 miles N.N.E. from the Manawatu Gorge. The Southern Ruahines present a narrow, high (1000-1220 m) wall, bounded in the west by the Pohangina Valley and farmland which extends to the foot of the main slopes (Elder, 1958).

Ecologically it is a well-defined area. The climate is influenced by the Manawatu Gorge which forms a minor wind funnel for the prevailing westerly winds and brings a high proportion of cloud to the lower slopes. The climatic influence is implicated in the plant associations of the region (Franklin, 1967). On either side of the Gorge similar forest and scrub patterns prevail. A number of species characteristic of the Range show a definite drop of about 150 m in the upper limits of their altitudinal range.

The rimu-rata forest (366-700 m) typically exhibits large emergent trees of both species with numerous understorey species, notably tawa (Beilschmeidia tawa) and supplejack (Rhipogonum scandens). Below 427 m supplejack thickets are virtually impenetrable. Above 427 m the forest is more open and large-leaved coprosmas (Coprosma lucida, C.grandifolia and C.tenuifolia), rangiora (Brachyglottis repanda) and pate (Schefflera digitata) are present. Isolated pure stands of black beech (Nothofagus solandri var. solandri) occur on exposed spurs. Throughout the area two climbing ratas (Metrosideros diffusa and M.perforata) abound. Unfortunately both species, especially the former, may be closely associated with *M.robusta*; this created some technical problems during sampling (see Section II2).

The area's recent biotic history has been greatly influenced by introduced animals. The liberation of pigs, goats, deer, cattle and opossums was followed by a recession of indigenous palatable plant species and their associated animals. Recorded opossum (*Trichosurus vulpecula*) liberations took place in Totara Reserve (1900) and Makawakawa (1918) (Elder, 1958). By 1958 they had established a high stable population up to 608 m in the Southern Ruahines. At present a N.Z. Forest Service exclosure plot, erected in 1970, shows no apparent contrast with the surrounding vegetation and the area appears to have reached an equilibrium which is probably maintained by organised hunting and trapping.

Adverse weather conditions have never been recorded as inflicting severe damage in the Southern Ruahines. The gale of 1936 affected only trees on exposed spurs. Snow damage is rare below 650 m (Elder, 1958). The area is usually fairly damp with an annual rainfall of about 2000 mm. However, droughts were recorded in the summers of 1945-46, 1969-70 and 1972-73. The drought of 1969-70 was severe enough to cause the death of some plant species in Bledisloe Park, Palmerston North (Atkinson & Greenwood, 1972), though the one rata present was recorded as being unaffected. The drought of 1972-73 was not as severe (Table I) but did come during the oviposition period in the life-cycle of *P.radicis* (see Section II3).

The soil of the area is described as skeletal, light-brown to yellow, silt loam derived from greywacke. It has a low natural fertility and a pH of approximately 5.6 (Taylor, 1954).

Summer of	Dec	Jan	Feb	Mar
1969-70	92.7 (13)	27.4 (8)	8.9 (6)	83.1 (15)
1972-73	40.4 (12)	52.4 (7)	16.5 (7)	102.3 (14)

TABLE 1 A COMPARISON OF DROUGHT CONDITIONS FOR PALMERSTON NORTH - MONTHLY RAINFALL (mm)

( ) = raindays, i.e. 0.1 mm rain

Data from Herbage Lab., D.S.I.R., Palmerston North

Materials and Methods

During the study air and soil temperatures and rainfall were recorded continually. Wind speed and soil moisture were measured during the flight period of P.radicis and dispersion studies respectively.

Rainfall during the study period was measured with a calibrated 15.2 cm diameter gauge sited in open farmland about 100 m from the bush margin (Fig. 2).

Minimum-maximum soil and air temperatures were recorded with two U-thermometers at a single site at approximately 450 m. Air temperatures were taken in the shade at a height of 1.5 m and soil temperatures were recorded at about 5 cm depth. During the flight period of P.radicis (February-March) further meteorological recordings were obtained by means of a chart thermohygrometer. Periodic anemometer checks were also made to determine wind velocity.

Results

The total rainfall for 1972 amounted to 1017 mm; the distribution can be seen in Fig.3

Soil and air temperatures reached a peak in February and their lowest level in June. Air temperatures were always more variable, and reached higher and lower extremes than soil temperatures. The constancy of conditions during the drought period are shown by a week's record from the thermohygrometer (Fig.4)

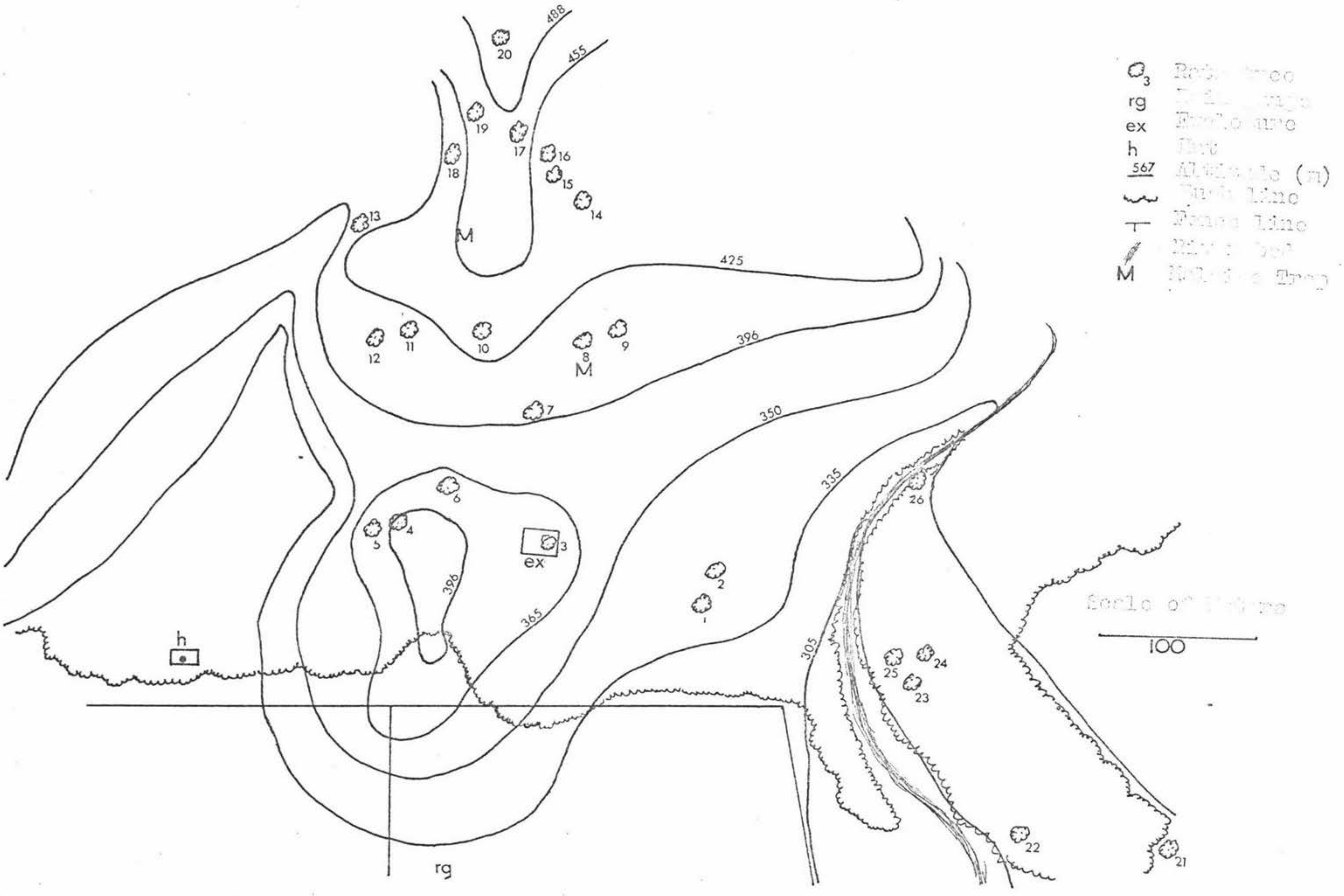


Fig.2 Detail of the study area- S.W. Ruahine Range.

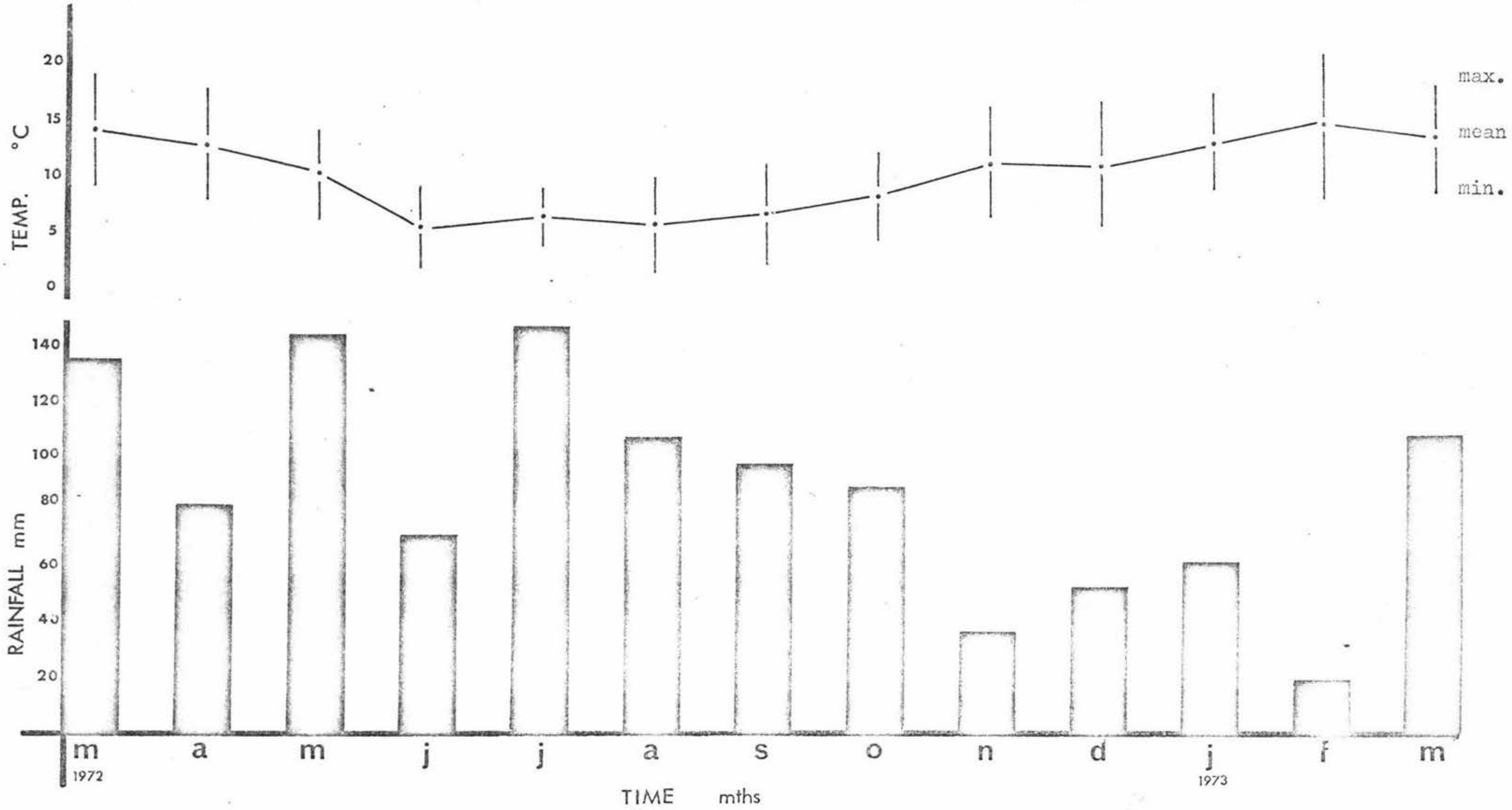


Fig.3 Meteorological records from the study area showing rainfall (mm) and min., max. and mean soil temperatures for each month from March 1972 - March 1973.

Fig .4 . Typical thermohydrograph record obtained at the study area during the 1975 flight period.

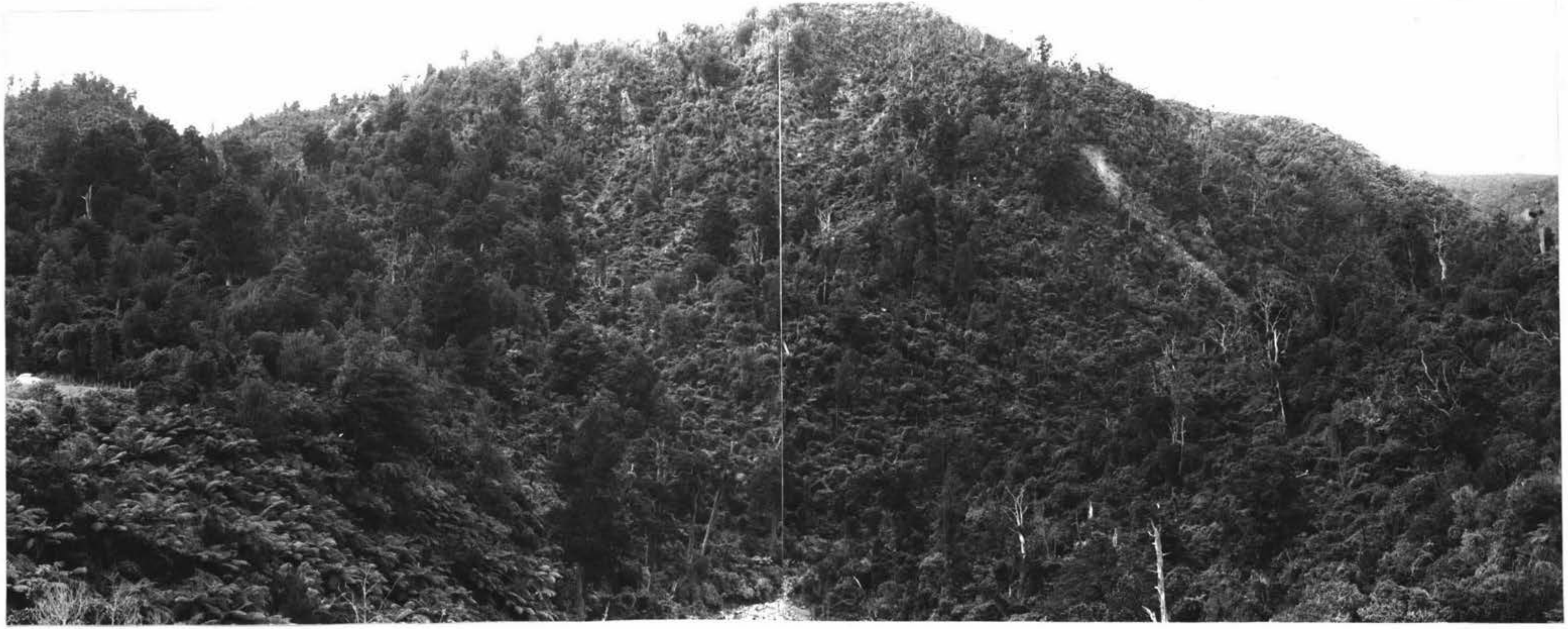


Plate 1 The study area - S.W.Ruahine Range - showing some rata mortality. (South-facing valley side on left)

The Host, Northern Rata (*Metrosideros robusta* A. Cunn MYRTACEAE)

Introduction

The role of the host in any host-parasite system is of major importance. A favourable host-parasite association is crucial for the establishment and development of the parasite. A high degree of phenological synchronisation and physiological compatability with the host is necessary for the success of the parasite.

Northern rata is usually found as an epiphyte, typically on rimu (*Dacrydium cupressinum*). The 'trunk' of the tree is built up of coalescing descending roots. The mature tree possesses a large umbrella-like canopy and a massive irregular trunk with a central cavity which is hollow or filled with the decaying trunk of the host. The tree is of little direct economic importance, so not surprisingly, little is known of the tree's phenology. The dubious value of what is known is aptly collated in Druce's 'Brer Rata and Uncle Rimus' (1972). This section strives to elucidate the many biological aspects of *Metrosideros robusta* which may be implicated in the host-parasite association with *Proterodiplosis radicis*.

Description

Metrosideros robusta is an endemic evergreen tree up to 25 m or more tall with a trunk approximately 2 m in diameter. The leaves are glabrous and elliptical in shape (25-50 mm long x 15-20 mm wide). The tree has a conspicuous, bright crimson, many-flowered, terminal inflorescence (Allan, 1961).

Distribution

M.robusta is distributed throughout the North Island and the north of the South Island of New Zealand. Typically it is a prominent species in lower montane forests. The tree's upper altitudinal limit in the

Southern Ruahine Range is about 760 m (Elder, 1958) and there is a marked lowering of this limit in the southern regions of the species' range (Franklin, 1967).

Relationships

Some 25 species of Metrosideros have been recorded from the Southern Hemisphere (Balgooy, 1963), and 11 of these are endemic to New Zealand. Superficially M.robusta resembles southern rata (M.umbellata) and the geographical and ecological ranges of the two species overlap to a large extent, although there has been no recorded hybridisation between the two. Cooper (1954,1958), on the other hand, has documented hybrids from M.robusta and pohutukawa (M.excelsa) and has produced evidence of large-scale hybrid introgression in some areas (see Section III 2).

The typical epiphytic growth form of M.robusta possibly represents an evolutionary intermediate between the climbers M.perforata, M.diffusa and M.tomentosa, and the true arborescent forms M.excelsa and M.umbellata.

History

Couper (1953) recorded Metrosideros pollen from the Eocene period, although no specific identification was made.

Morphological Features

a. The root system

From observations of both standing and fallen trees it was evident that epiphytic ratas had a shallow rooting system. Upon reaching the ground the roots descending the host's trunk may radiate 20 m or more from the trunk and are readily traceable over most of their length. This shallow rooting system is probably the result of the epiphytic growth form, as the epiphyte is initially supported by the host tree and has no need of deep tap roots. By the time the host's trunk has decayed the rata is a massive structure supported by an extensive root system.

Three root types are recognisable in the secondary rooting system. On the 'trunk' and older branches 'adventitious' roots grow closely adhered to the bark. These aerial roots are thickened towards the apex (ca.3 mm diameter), reddish in colour and devoid of root hairs. On the ground similar roots, creamy-white in colour, act as 'runners' from damaged main roots or between beds of fine (0.2 mm diameter) branched rootlets. These fine rootlets, upon which root hairs (ca.0.5 mm in length) may be present, are generally restricted to rotting wood and rich humus layers of the soil. It is these roots which are galled by Proterodiplosis radicis (Plate 2). Other root abnormalities have been found on M.robusta and Beddie (1953) recalls Percy's finding (1928) of root swellings "of considerable size" on young terrestial seedlings of rata. No pathological agent could be associated with the swellings and they were considered to be homologous to the lignotubers of Eucalyptus.

b. The shoot system

The terminal growing point of the vegetative shoot aborts after its seasonal growth is completed so that the shoot terminates with a pair of buds, axillary to the uppermost leaves. Branching therefore is usually dichotomous although some deviation from this pattern is found in seedlings and cuttings. A brilliant crimson inflorescence may develop from the axillary bud pair. The primary axes of the inflorescence terminate in a vegetative bud which may later develop into a leafy branch (Dawson, 1968).

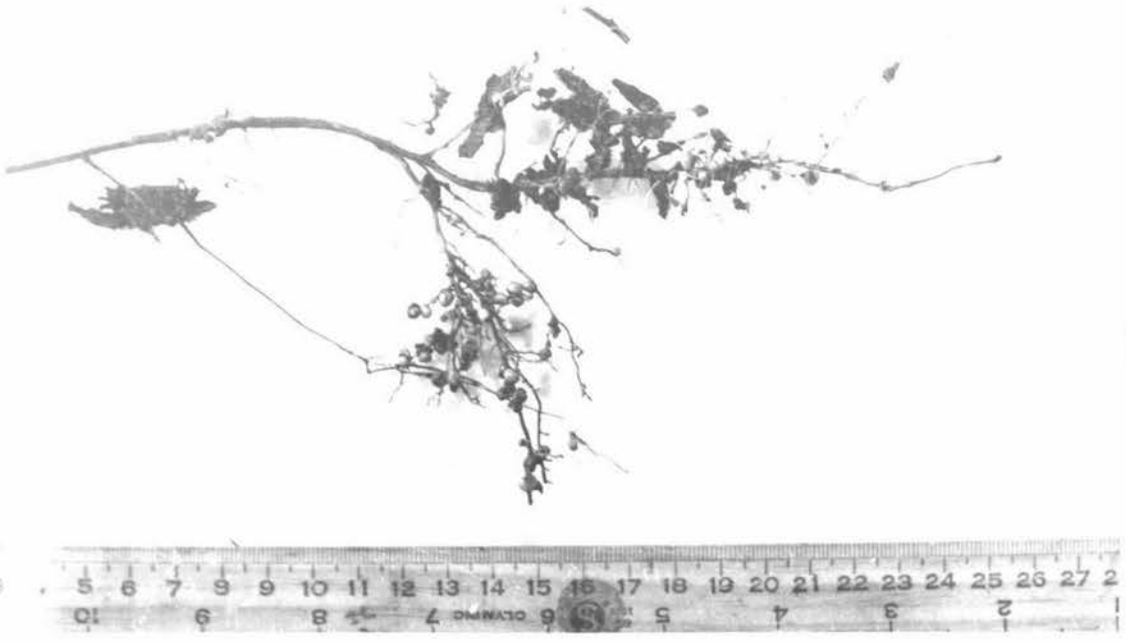


Plate 2 Galls of P.radicis onthe fine rootlets of M.robusta.

Plate 3 The exposed , white, 'runner' roots of M. robusta : note also the presence of M. diffusa.

Plate 4 The typical growth-form of Metrosideros robusta. The roots of a young rata are descending the trunk of a large rimu (scale = 17 cm - increment band also in place).

Anatomy

The anatomy of the fine rootlets is discussed in Section III.2 Suffice to say the root has a tetrach stele with a distinct endodermis and asymmetrically thickened epidermal cells.

The leaves show some xeric features, being relatively thick and leathery and possessing a thick shiny cuticle. Further leaf anatomy is described by Betts (1919).

Trunk growth rings are not readily discernible as large vessels are evenly distributed throughout the ring. The annual nature of any ring formation may be further hidden by the irregular nature of trunk growth; the possibility of severe defoliation by insects and opossums (Trichosurus vulpecula), and the occurrence of minor spring flushes in trees at lower altitudes. These factors could contribute to false ring formation and result in an overestimate of age or an underestimate of seasonal biomass production (Glock & Agerter, 1963).

Chemistry

Largely because of its close relationship with Eucalyptus, the chemistry of M.robusta has been reasonably well studied (Table 2). The major secondary plant compounds are phenolics, with a notable absence of alkaloids and saponins. Secondary plant compounds, especially phenolic compounds, have been implicated in resistance to pathogens and insects in a number of plants (Fraenkel, 1967; Miles, 1969).

Compound	Reference
Leaf tannins	Betts, 1919
Bark tannins	Aston, 1918
Leucoanthocyanins	Cambie *et al.*, 1965
Triterpenes	"
Essential oils	"
	Gardner, 1931
	Brooker, 1963

TABLE 2 THE CHEMISTRY OF *M.robusta*

Organism			Reference
Fungi			Gilmour, 1966
	Saprophytes		Cunningham, 1963, 1965
INSECTA			
Homoptera	Coccoididae	6 spp.	Hoy, 1958
Coleoptera	Scolytidae	1 sp.	
	Pyrallidae	1 sp.	
Phasmidia		1 sp.	Meads, pers. comm., 1972
Homoptera	Psyllidae probably *Trioza*		Valentine, pers.comm., 1972
Diptera	Cecidiomyiidae *P.radicis*		this study
VERTEBRATA	*Trichosurus vulpecula*		Manson, 1958

TABLE 3 DEBILITATING ORGANISMS RECORDED FROM *Metrosideros robusta*

The Phenology of Northern Rata

The phenology of M.robusta, beyond its conspicuous summer flowering period, has not previously been documented in any detail, although the closely related M.umbellata has recently been extensively studied (Wardle, 1971). The importance of phenological synchronisation in a host-parasite association warrants a close investigation of the host's phenology in any study. The intimate association of P.radicis with its host made such a study imperative.

Materials and Methods

All phenological studies, unless otherwise stated, took place in the study area. General field observations of growth flushes, leaf fall etc. were supplemented by glasshouse propagations of seedlings and cuttings, measurement of shoot and root growth, and increment banding of a number of trees.

Seedlings were germinated on damp filter paper or peat and later re-potted in a sand-peat mixture and grown in the glasshouse. Cuttings were obtained in spring and later summer from trees 6 and 8 and grown in a sand-peat mixture, under mist, in the glasshouse.

Twenty shoots of the lower canopy of tree 6 were tagged and measured periodically from immediately after bud break to the cessation of growth.

The growth of the fine rootlets proved difficult to measure without their eventual destruction. A number of methods were compared and finally a semi-objective method, in which field observations were supplemented with laboratory counts of root tips per unit root weight, was selected. Five small samples of rootlets were taken from one tree chosen a random during normal visits to the study area from November 1972 to March 1973.

The number of root tips per sample was counted under a dissecting microscope. The sample was then weighed and a 'rooting index' was calculated by dividing the number of root tips by the weight of the sample. It is obvious that this index does not necessarily represent root biomass as it does not consider root extension; however, it does provide an estimate of the available sites for gall formation, which is relevant to the overall study.

Increment bands were made from lengths of 2 cm steel packing tape and 10 x 0.7 cm extension springs to suit individual trees. Eight trees (3,4,6,7,8,13,14,15) encompassing a range of diameters and altitudes were banded at breast height in May 1972. The bands were allowed to 'settle' for two weeks before being marked. The movement of the mark was measured periodically with callipers.

Results

The commencement and duration of phenological events are presented in Table 4. Data for shoot, root and increment growth can be found in Appendix 1. The phenology of M.robusta, in terms of the commitment of biomass, is illustrated in Fig.5

Discussion

The onset of flowering in individual trees varied considerably within the relatively small altitudinal range present in the area. In most instances trees at lower altitudes commenced flowering in late December while those at higher altitudes flowered one to three weeks later. The duration of flowering was two to three weeks. Mature seed was collected from fallen capsules in August.

Seeds were found to germinate readily on damp filter paper or peat and seedlings when potted grew continuously in the glasshouse to attain a height of 12 cm in nine months. The ease of growing seedlings contrasts

Plate 5 An increment band used to detect cambial growth.

Phenological activity	commencement	termination	duration
Spring shoot growth	late September		2-3 weeks
Flowering	late December	mid-June	2-3 weeks
Summer	mid-June	mid-August	16 weeks
Seed fall	August?		?
Leaf fall	December	June?	continuous?
Root growth	October	May?	continuous
Increment growth	very slow		?

TABLE 4 PHENOLOGICAL DATA FOR *Metrosideros robusta*

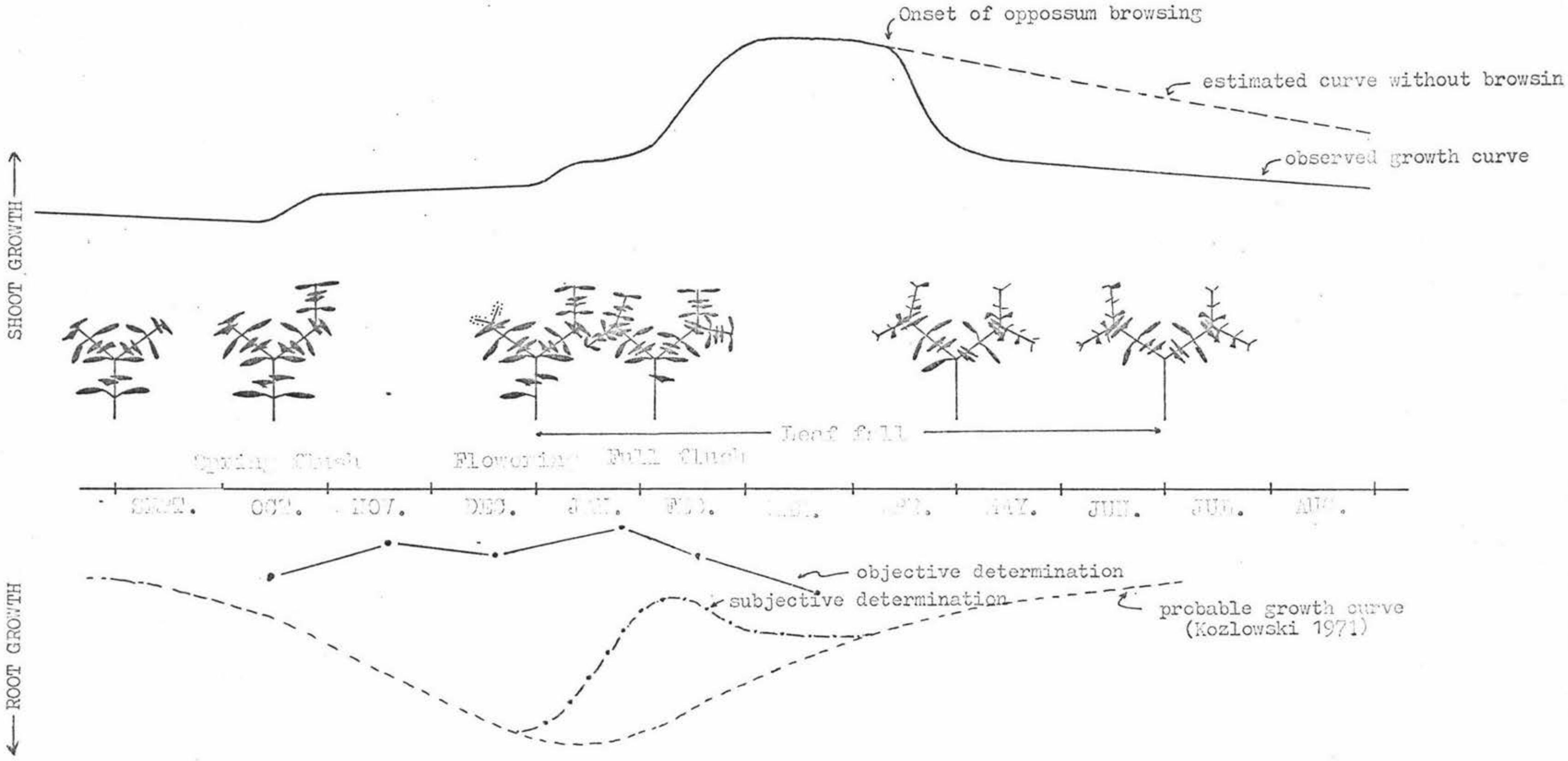


Fig. 5 The phenology of Metrosideros robusta 1972-1973 .

markedly with the dearth of seedlings in the forest. Terrestial seedlings were never found in the study area or any other area sampled during this investigation.

Cuttings were more difficult to establish and required a very damp rooting medium. Cuttings remained dormant until the following spring but once established they grew vigorously in the glasshouse. Late summer cuttings were more successful than spring cuttings.

The main shoot flush occurs immediately after flowering. The growth of tagged shoots was greater in the six weeks following bud break (Appendix I Table A). Shoot growth ceased in May. A minor spring flush was observed in a number of trees below 400 m (trees 1,2,3, 4,5,22,23,24,25,26). Thus flush, however, involved a comparatively small number of shoots and these were localised within individual canopies.

Root growth for most trees is a difficult parameter to assess, as exposed roots rapidly dessicate making sequential observations impossible. The results of laboratory counts of root tips and the subsequent root index are rather erratic (Appendix I). Subjectively there appeared to be an increase in root growth during spring and autumn. The 'root index' did not reveal the obvious effects of the 1972-73 summer drought in which many root buds were desiccated. Kozlowski (1971) states that root growth of temperate zone trees usually begins earlier in spring and continues for a longer period than shoot growth, although typically roots show greater variability in growth rates than shoots. The observed root index, the subjective observations and the probably root growth patterns (Kozlowski, 1971) are plotted in Fig. 5

Increment bands have been criticised for their indistinct recording of the initiation of seasonal growth (Kozlowski, 1971). However, only a gross annual measurement was required here to be correlated to the degree of infestation of <u>P.radicis</u>. After a year's growth no significant increment gain could be recorded even though some trees exceeded 5 m in circumference (Appendix I). This suggests a very slow rate of growth: however, the irregular growth form of M.robusta may obscure moderate, but localised, growth.

From the seasonal nature of shoot growth and the dichotomous branching habit of M.robusta it was estimated that leaves were retained for about three years. A number of leaves were present on the primary twigs at the beginning of the third season. Leaf fall appeared to be continuous although a reddening of senescing leaves, and a marked increase in leaf fall, was noted from November to April.

The overall pattern of the seasonal allocation of biomass is summed subjectively in Fig.5

Rata Mortality

Several reasons for the mortality of M.robusta were suggested in the Westland Interdepartmental Report (NZFS, 1955). These hypotheses could be broadly divided into climatic and biotic arguments. There was some evidence that climatic factors could cause localised mortality. Cockayne (1923) reported wind as the probable cause of death of M.robusta on Kapiti Island. However, the widespread mortality appeared to be closely related to the introduction and increase of exotic animals. It was generally conceded that the opossum was the main suspect either in initiating or achieving the mortality of rata (Holloway, Hoy et al., 1955, NZFS).

Hoy (1955,1958) assessed the mortality of M.robusta in the Southern Ruahines and reported that "in the majority of areas where the death of rata is occurring there has been a general thinning of the canopy and a considerable loss of surface litter". He also stated that, although the proportion of dead trees to total trees was low, the remaining trees were in an 'unhealthy' state. He observed that mortality occurred in ratas of all sizes and appeared to be unrelated to topography. It was obvious that an objective investigation of the mortality of M.robusta would be of some benefit in assessing the role of P.radicis in the problem.

Materials and Methods

Four valley sides (two north- and two south-facing) in the study area were viewed through a telescope from various vantage points. The number of standing rata, both dead and alive, was recorded and the dead trees classed as 'young', 'medium' or 'old' depending on their trunk diameters. Typically 'old' dead trees had completely overcome their host before dying; 'medium' dead trees had encircled most of the host's trunk while 'young' dead trees had only one or two narrow descending roots.

Plate 9 Examples of dead rata (right) within the study area; note the thin canopy of the tree on the left.

Results

The results of counts of dead and live M.robusta in the study area are expressed in Table V. Rata mortality is expressed as age-class percentages in Fig.6

Discussion

The results show that in fact 50% of the standing M.robusta is dead (90 observations). This mortality occurred in trees of all sizes, and presumably all ages; however, more than 70% occurred in large old trees. The difference between mortality on the northern and southern slopes was not significant (t = 0.322; 1 degree freedom).

A number of detrimental agents have been recorded from M.robusta and are recorded in Table III. The overriding impression when studying M.robusta is its susceptibility to attacks of all kind. Anoplaspis metrosideri was recorded by Hoy (1958) as the most important coccid, causing localised mortality in the Southern Ruahines. In the study area trees 6 and 8 were badly infested. Trioza sp. was observed in the study area attacking young shoots immediately after bud break. Severely attacked leaves were dimpled and the shoots stunted.

The flowers and young leaves of M.robusta are a major source of food for the opossum (Manson, 1958). Families of opossum congregate on a tree and browse as long as fresh shoots persist, leaving neighbouring trees completely untouched (Wardle, 1971). In a harsh winter, opossums feed more voraciously and mature leaves may be eaten (Meads, pers. comm.). In the study area, opossum attack coincided with the onset of winter(May), rather than the onset of shoot growth (January). In lightly browsed trees the damage was restricted to the top half of the young leaves. The timing and nature of the attack suggested that although M.robusta is a staple food in some areas (Manson, 1958), it was not necessarily a preferred food in the study area.

ASPECT	STATE OF TREE			
	ALIVE	DEAD		
		Old	Medium	Young
N	11	13	3	3
S	12	6	1	1
N	21	11	–	–
S	1	3	3	1
Total	45	33	7	5

TABLE V OBSERVATIONS ON TREE HEALTH OF RATA IN THE STUDY AREA

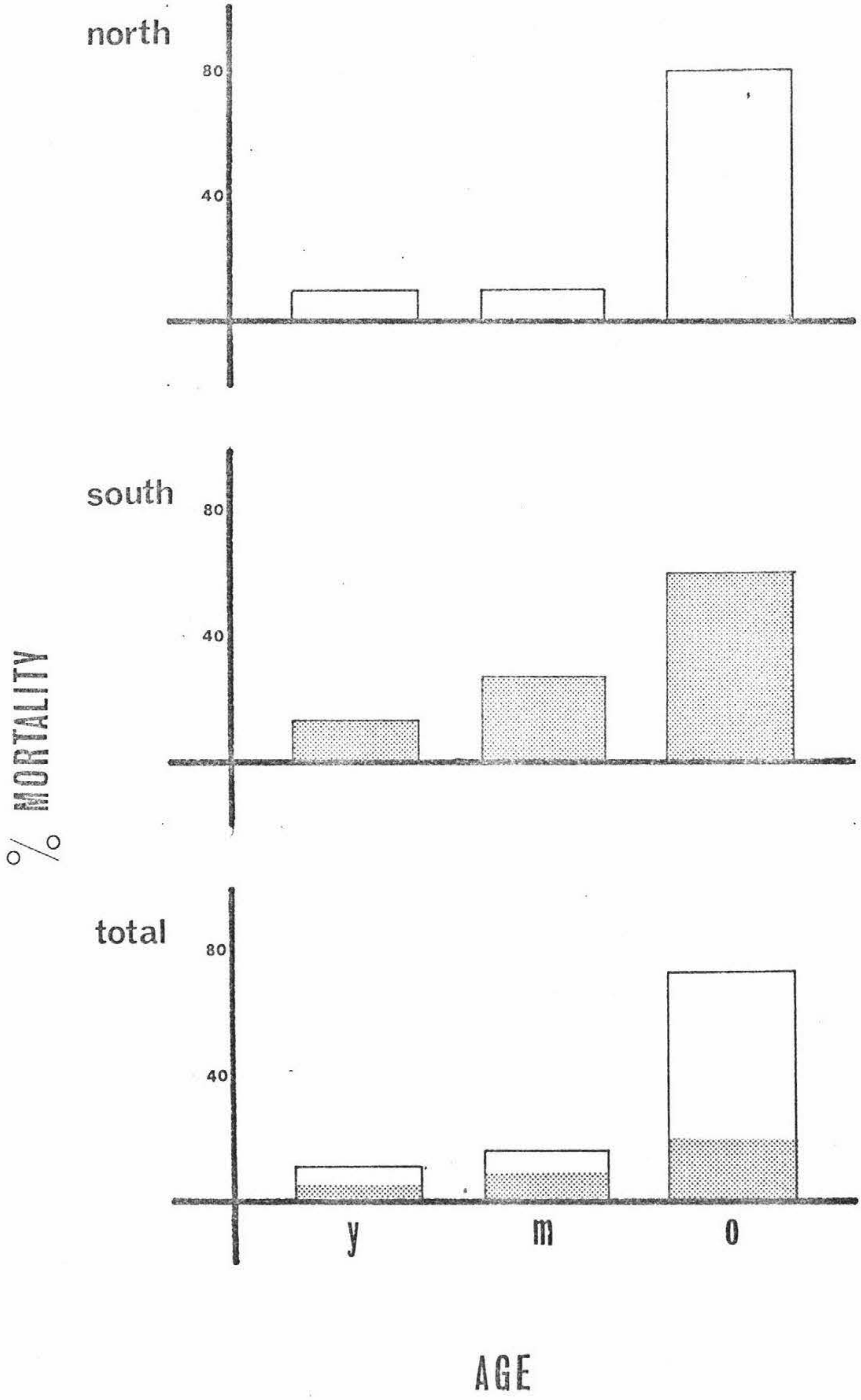


Fig. 6 The mortality of rata in the study area expressed as age-class percentages on a, North-facing slopes b, South-facing slopes c, total.
y = young trees m = medium-aged trees o= old trees.

Climatic damage to M.robusta in the study area did not appear to be great. Only a small number of trees which had been blown over were observed. One young tree had been snapped, with its host, half-way along its length. Wind did accentuate scolytid attack in the study area. After a strong wind, branches up to 10 cm diameter were often found near large exposed trees. Many of these branches, although still carrying green leaves, were ring-barked to some extent at the point of separation from the tree.

Some concern was expressed in the 1955 Report that the thinning of the understorey vegetation and the compacting of the soil by introduced animals could affect root growth of some trees. Some root desiccation was observed in the study area during the summer drought (1972-73); however, two mature trees growing in exposed, grazed pasture adjacent to the forest had the appearance of being the healthiest trees encountered during this two-year study.

The Description of Proterodiplosis radicis Wyatt

Introduction

The Cecidomyiidae are generally easily distinguished from other Diptera by a number of characters. Their holoptic eyes and short coxae; the absence of tibial spurs, reduced wing venation and their elaborate antennal sensoria separate them from their nearest relatives, the Mycetophilidae. Within the family, however, the Cecidomyiidae possess few striking morphological characters of use to the systematist. The adults are small, delicate flies with long antennae and legs. Differences in antennal structure are used by systematists at all taxonomic levels. The males of many species have antennae with intricately whorled sensoria. The wings, however, have few longitudinal veins and no obvious cross veins. The larvae are rather stout and often brightly coloured with a characteristic dorsal ocular stigma in the first or second thoracic segment. The small, incompletely differentiated head is followed by a supernumerary segment, three thoracic segments and nine abdominal segments. The most characteristic larval structure, usually associated with the final instar, is an elongated toothed sclerite, or sternal spatula, situated mid-ventrally on the thorax (Felt, 1925).

Early identifications stressed the host plant and gall structure rather than comparative insect morphology. There was a propensity to describe a gall midge from an unrecorded gall as a new species. At present the number of genera containing only one species is rather high (Mani, 1964), indicating that either many more species await discovery or, alternatively, that many genera are invalid. The cecid fauna of New Zealand is probably a good example of the former, while the revisions of Harris (1966) and Edwards & Pritchard (1968) show that the latter is the case in some families.

The structural similarity of the cecids makes their identification difficult. Barnes (1953) proposed that biological as well as morphological data should be utilised for the identification of species. His monograph (Barnes 1946,1948,1949,1951,1956) on the economically important gall midges contains detailed information on distribution, life-cycle, host range, parasites etc. to aid identification. Recently, however, Harris (1968) has made a more detailed examination of comparative morphology and has incorporated in his identifications such characters as the curvature of wing veins, chaetotoxy of the thorax and abdomen, and many larval characters previously ignored. The larval characters used to distinguish between genera are the number and arrangement of papillae and setae, and the forms of the sternal spatula, if present. With this in mind Wyatt's (1963) description of P.radicis has been extended and characteristics of the egg, and the first and second larval instars, included.

Materials and Methods

All specimens were obtained from rata root material collected from the study area. Characters and terminology used follow that of Harris (1966).

Preparations of whole specimens mounted upon microscope slides could readily be made of all developmental stages, although adults were usually mounted separately. Larvae, pupae and adults were cleared when necessary with 10% KOH. Specimens were mounted permanently in Canada balsam or temporarily in glycerine. Temporary mounts could subsequently be made semi-permanent by ringing with a proprietary, ethyl-acetate-based, clear varnish ("Cutex"). Specimens were viewed and measured under normal and phase contrast, light, microscopes. Photographs were taken with Ilford FP4 film.

Head capsule measurements of larvae were taken at the broadest region. Length (between the most anterior portion of the head and the most posterior point of the abdomen), and breadth (at the widest point of the abdomen) were also measured for both larvae and pupae. Adults were measured from the head to the tip of the genitalia, and from wing tip to wing tip (with the wings in an extended position).

Results

Gross body measurements are collated in Table VI

The egg

The egg is elliptical with a semi-transparent, shiny chorion. When newly laid it is translucent with a central band of pale yellow fat droplets. Prior to hatching both the pale yellow fat-body and the ocular stigma of the larva are discernable.

The larvae

Three larval instars could be determined by head capsule measurements (Table VII Fig.7). Each instar was readily identifiable by its size, colouration, and the presence or absence of characteristic appendages. All stages had a similar head capsule, with well developed antennae, and their dorsal and lateral surfaces covered to some extent with small scale-like papillae.

The first instar

The first larval instar has a small amount of pale yellow fat body laterally and a brighter, centrally situated, yellow-orange fat globule. This instar has no sternal spatula but is characterised by a pair of curved posterior processes on the terminal segment (Fig.8). These are effectively used as levers for locomotion when they are tucked under the abdomen to grip the substrate. The small dorsal scale-like papillae are

Developmental stage	No. of individuals measured	Range of lengths (mm)	Range of widths (mm)
Egg	5	0.280	0.112
First larval instar	38	0.37-0.45	0.15-0.25
Second larval instar*	100	0.66-1.38	0.18-0.78
Third larval instar	100	1.35-3.51**	0.6-1.08**
Pupa ♂	5	2.70	0.70
♀	5	3.65	1.20
Adult ♂	5	1.43	3.84
♀	5	3.85	4.05

TABLE VI GROSS BODY MEASUREMENTS OF P. radicis

* A great range in size in these stages denotes major feeding periods.

** Overlapping of measurements probably the manifestation of sex differences - female larvae being larger than male larvae when fully developed.

Larval instar	Head capsule breadth
1	12 m ±0.5 (.03 m ±0.002)
2	17 m ±0.5 (.043 m ±0.002)
3	23 m ±0.5 (.058 m ±0.002)

TABLE VII AVERAGE HEAD CAPSULE BREADTHS OF THE 3 LARVAL INSTARS

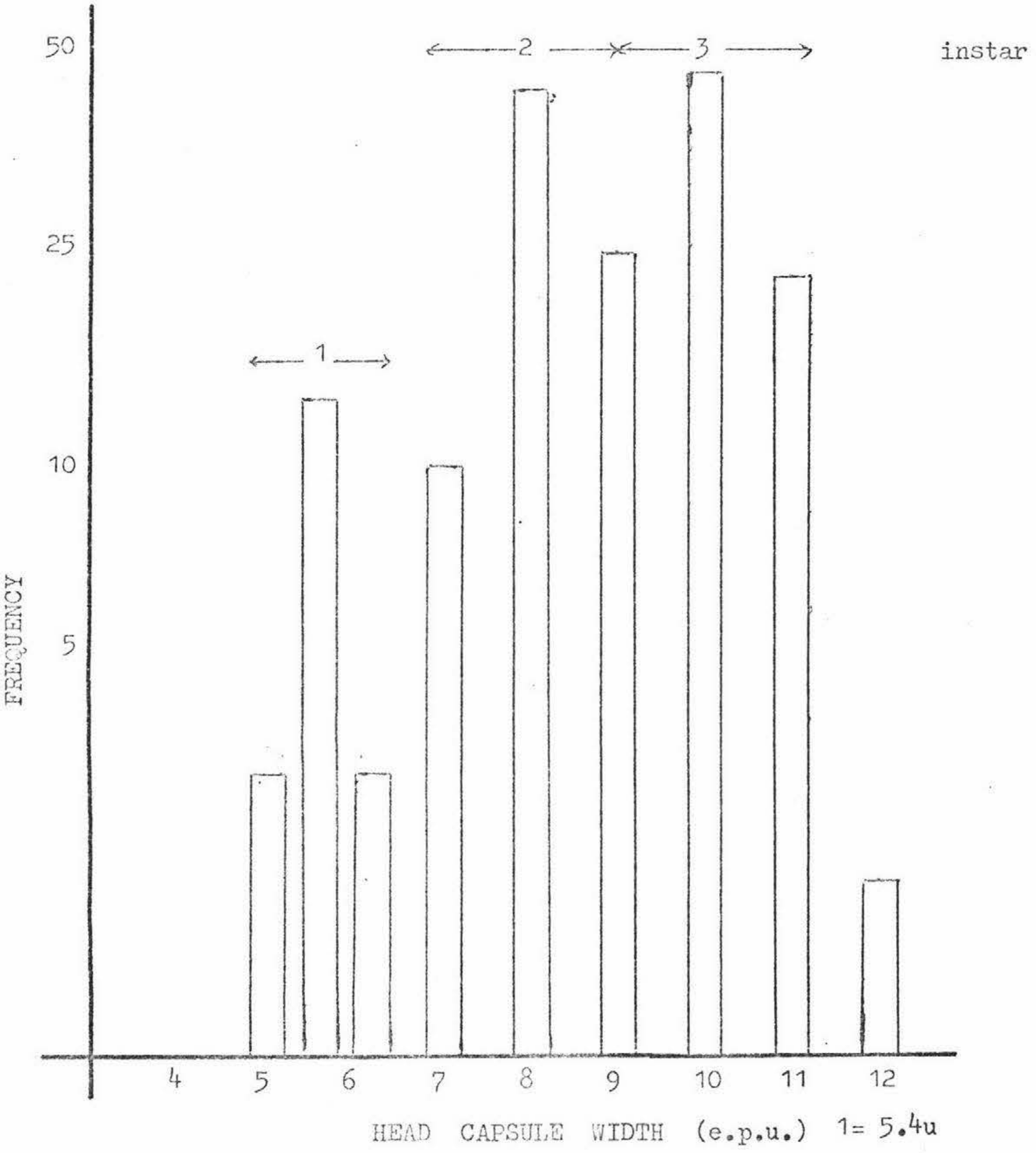


Fig.7 Histogram of head capsule widths of the larvae of P. radicis.

supplemented on each segment, excluding the last, with a row of larger papillae which extend to the lateral edge. Those of the collar segment are slightly smaller and centrally grouped. Ventrally the segments are smooth apart from three to four rows of minute spinules.

The terminal segment bears a ventral elliptical anus and four ventral papillae between the caudal processes. The penultimate caudal segment bears the only functional pair of spiracles which are situated on raised dorsal prominences.

The second instar

The second larval instar is peripneustic with a well-developed tracheal system clearly visible. The larvae are pale yellow and lack prominent appendages or papillae. There is no sternal spatula and the arrangement of papillae is virtually identical to that of the first instar (Fig 8).

The third instar

The third larval instar has been described by Wyatt (1963). The larvae is also peripneustic and possesses a characteristic mid-ventral sternal spatula on the prothoracic segment. The body colour, however, is not 'yellowish' (Wyatt, 1963) but bright orange - a discrepancy which probably arose as Wyatt was dealing with preserved specimens. The transition from yellow to orange takes place soon after entering the third stadium. The change is initiated terminally and is soon complete. The ecological significance is not obvious - it may be due to change in the composition of the diet. The pupae and adults retain this orange colouration.

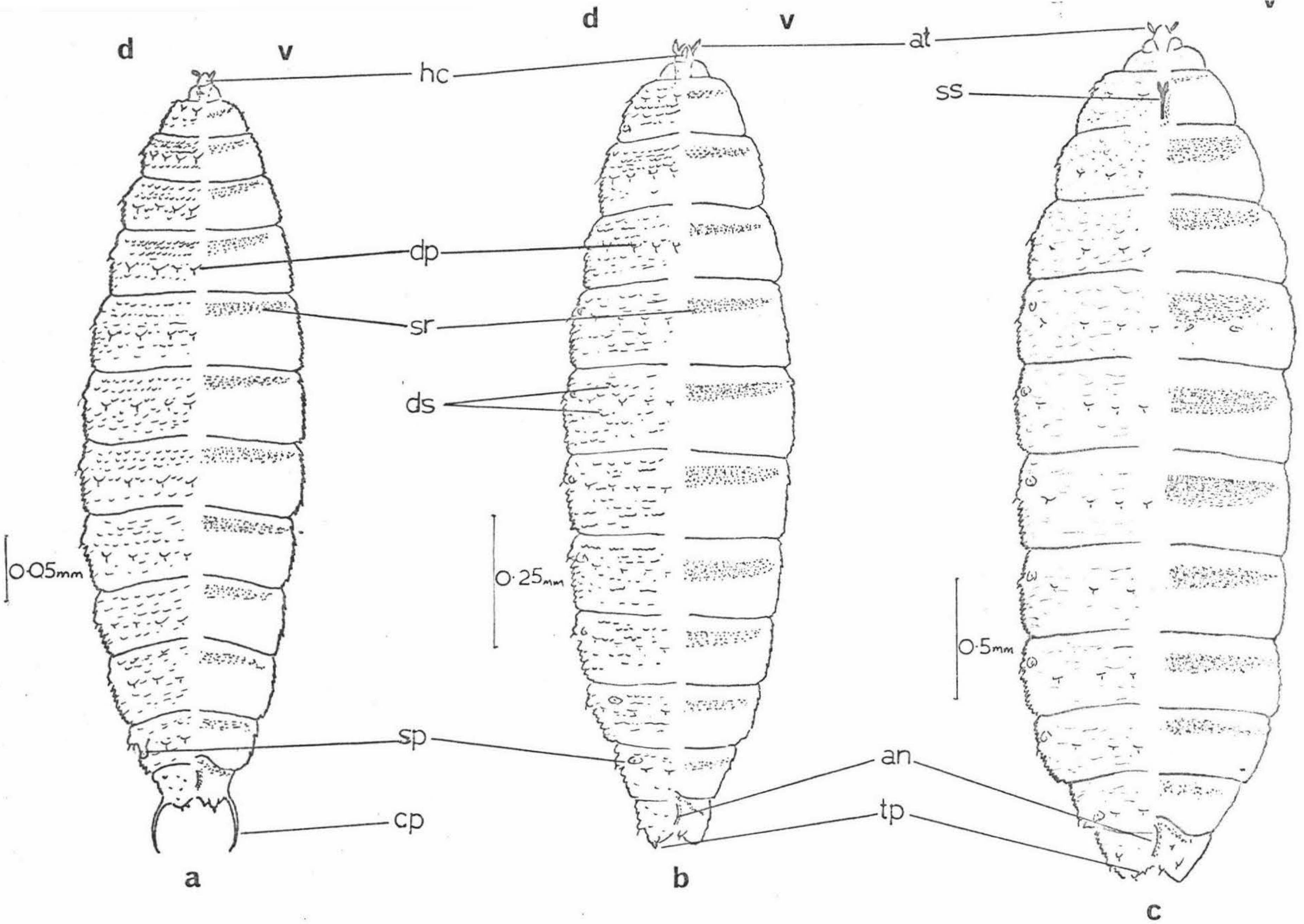


Fig.8 The appearance (from left to right) of the first, second and third instar larvae of *P. radicis*. (dorsal/ventral views) an, anus; at, antennae; cp, caudal process; dp, dorsal papillae; ds, dorsal spines; hc, head capsule; sp, spiracles; sr, spinule rows; ss, sternal spatula; tp, terminal papillae.

The pupae

Pupal morphology has rarely engendered much investigation. When pupae are present it is generally true that adult specimens, which afford more useful diagnostic characters, will also be available.

Prior to pupation the larva becomes distended and pales anteriorly. The dorsal ocular rtigma divides and the two portions migrate ventrally to lie at the base of the developing compound eye. During pupation the pigment multiplies to cover the whole eye. The head and thorax, with their appendages, blacken with maturity while the abdomen remains a pale orange colour.

A pair of acuminate, curved thoracic 'horns' protrude outward and forward from the prothorax and the head is crowned by a pair of short cephalic horns. The anterior dorsal edge of the abdominal segments bear small posteriorly directed papillae which presumably aid movement within the gall. The sex of the pupae can be determined by its size (TableVIII) and the structure of the genitalia and antennal segments.

The adults

The adults have been described in detail by Wyatt (1963). They are small fragile flies (Fig.9). Apart from obvious differences in size and genitalia, the sexes are readily differentiated by antennal structure. The antennae of the female are shorter with the segments having single nodes and lacking the elaborate sensoria of the male (Fig 10).

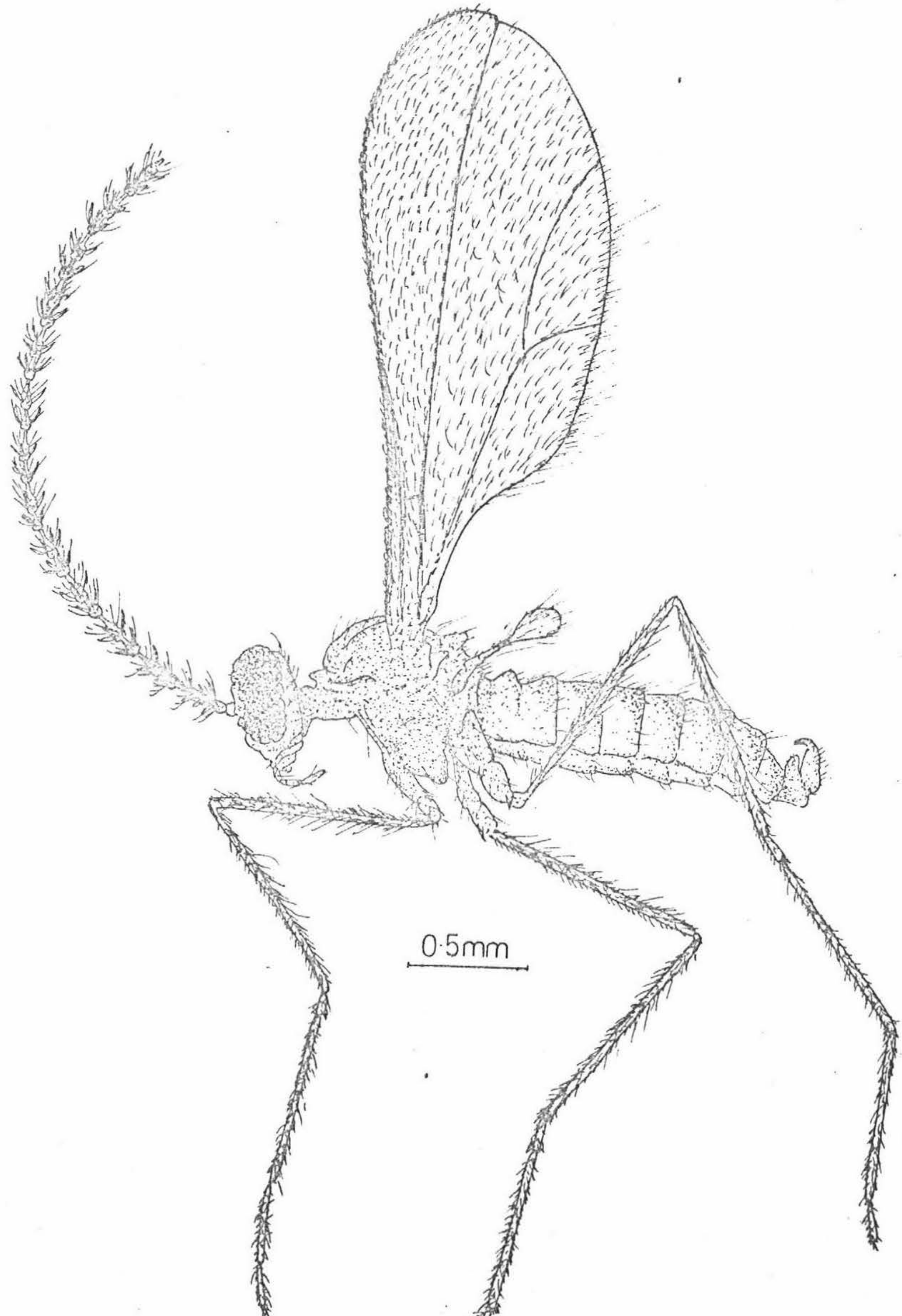


Fig. 9 Proterodiplosis radicis. Adult male. lateral view.

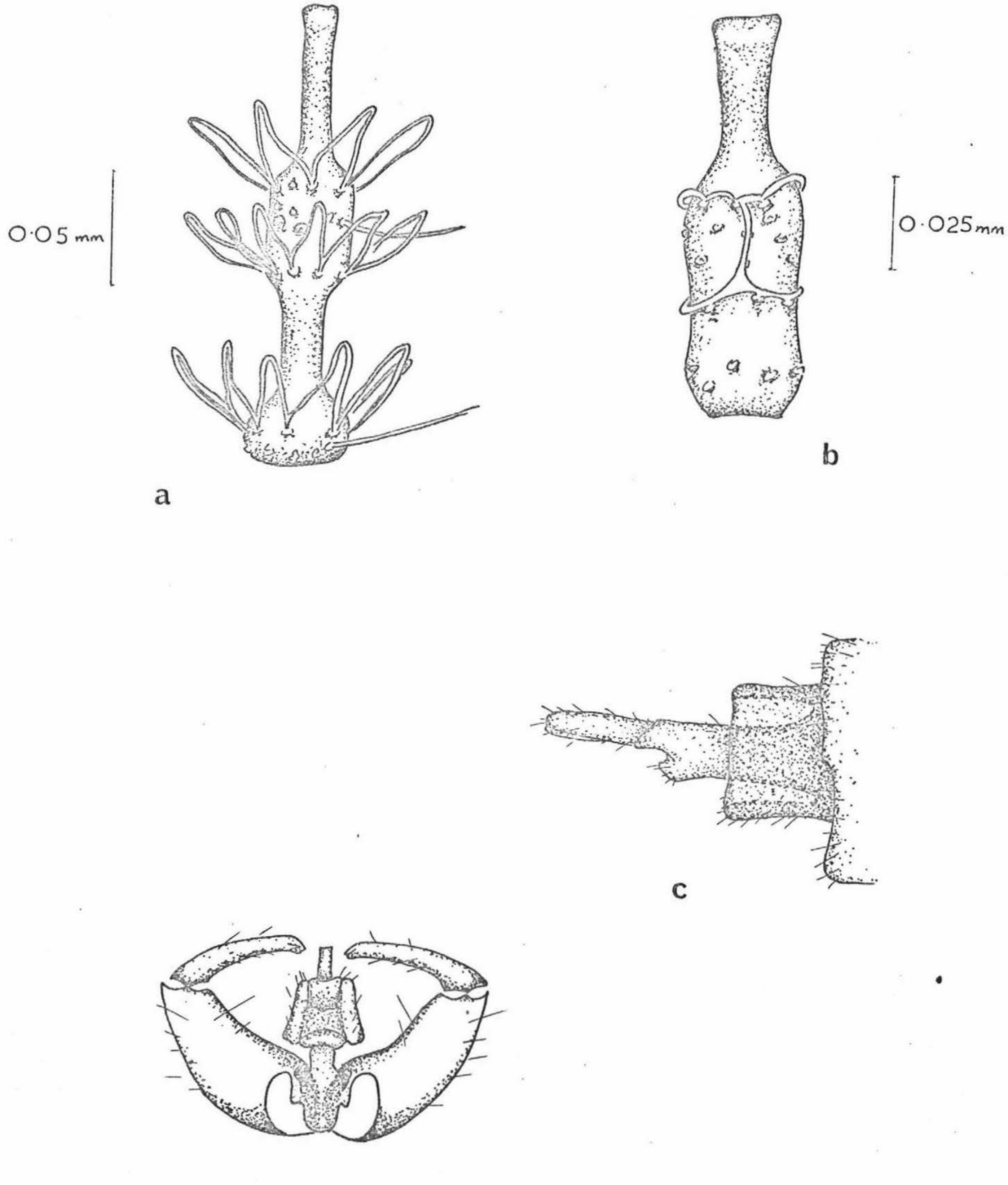


Fig. 10 Characters of sexual differentiation in P. radicis.
a, male antennal segment. b, female antennal segment
c, female genitalia. d, male genitalia.

THE BIOLOGY OF PROTERODIPLOSIS RADICIS

Introduction

Ecologically the Cecidomyiidae is a very diverse family and the full spectrum of feeding habits can be found among its members. However, the majority of the family are phytophagous and may form conspicuous plant galls during larval development. The adult gall midges usually do not feed and represent a short-lived reproductive and dispersal stage in the life cycle.

The galls induced by cecids exhibit a great diversity of form and structure and arise on a wide variety of plants. All parts of the plant are liable to attack; however, leaf galls are the most common. Typically the meristematic region of the plant is attacked and normal growth is modified to enclose the feeding larvae in a nutritive cavity (Mani, 1964). The type of gall formed is generally very specific for the particular midge and host plant involved, and may vary from a simple callus formation to a complex, distinct organ, incorporating characteristically differentiated gall tissue.

Such a close association with a host plant requires a close phenological synchrony, and gall midges tend to possess an individualistic ecology which suits their particular life style. Of necessity the growth of the larva is intimately associated with the development of the gall. This may be rapid, as in many leaf and flower bud galls: Felt (1925) records a life cycle of 9 to 11 days' duration for Contarinia sangricola. Typically, however, gall midges have only one generation per year. The duration of the adult stage is extremely short and can be measured in hours. The midges lay their eggs on, or in, the host plant's tissue and the ensuing larval development induces a localised abnormal growth on the plant.

The oviposition period coincides with the meristematic activity of the plant organ to be attacked and gall and larval development are often completed by winter. The midges usually over-winter as larvae within the gall. The pupation period is often short, although some species do over-winter as pupae (Felt, 1925). Pupation may take place within the gall or, more commonly, the final larval instar leaves the gall to pupate in a cocoon in the soil. An emergence exit is cut by the larva or the pupa and the pupa wriggles part the way out prior to adult emergence.

Within the gall the larva often exists in a semi-fluid environment and possibly feeds by absorption (Mani, 1964). The exact feeding mechanism is unknown. Mouth parts are minute and a possible lacerating organ, the sternal spatula (when present), is only developed in the final larval instar. Rafai _et al_. (1956) applied sensitive acoustic and photographic equipment close to the feeding larvae of the Hessian fly (_Mayetiola destructor_) and correlated rhythmic sucking noises with the application of the head region of the larvae to the plant. They suggested that phytophagous cecids feed on plant juices in this manner. There are no signs of excreta so commonly found in the galls of other Diptera, Lepidoptera and Coleoptera (Mani, 1964).

Within the gall the insect is relatively immune from normal regulatory processes, although the adults, eggs, and early instars are exposed and vulnerable, to some extent, to adverse weather conditions, predators and parasites. Common predators are other cecids, while a number of hymenopterous parasites, particularly Platygasteridae, have been reared from gall midges. Some midges, directly or indirectly, fall prey to various gall-eating vertebrates such as birds and rodents. However, the plasticity of the family is again evident in the reproductive processes encountered. Parthogenesis, larval and pupal paedogenesis and ovovivipary have been recorded in a number of different species (Felt, 1925, 1940: Mani, 1964; Harris, 1968), and the characteristically high biotic potential encountered in the family ensure its continuance (Hedlin & Johnson, 1963).

Current concepts of insect abundance and population dynamics attempt to explain the forces which regulate populations to achieve their often-observed numerical stability. However, the mode of action of the regularity forces is a matter of some debate. Nicholson (1933,1957,etc.) suggests that populations exist in a state of balance with their environment as a result of negating density-dependent processes, such as competition and production. Andrewartha and Birch (1954,1960), however, regard such density dependent processes as secondary to the intrinsic favourableness of the environment, and the capacity of the organism to exploit the favourable conditions. Milne (1957,1962) to some extent combines the two opposing views, while genetic regulation is emphasised by Chitty (1960) and Pimentel (1961). Chitty suggests a density-dependent genetic regulation where individual viability is inversely related to population density. Pimentel, on the other hand, states that the omnipotent regulator of populations is a continuous co-adaptation between trophic levels.

Despite the apparent differences in opinions, most would agree that the theories merely represent different evaluations of similar parameters, namely weather, food and shelter, competition, predation and parasitism, and genetic variability. These factors may influence the natality, mortality and dispersal of a population and thus regulate it. The degree of regulation effected by a particular parameter would depend on the population and its environment.

The Cecidomyiidae possess a number of interesting and often unique biological facets. The full extent of the family's plasticity is not known as few intensive ecological studies have been attempted. This section strives to elucidate the life cycle of P.radicis and the various regulatory factors influencing it.

The Sampling Programme

In the majority of ecological investigations the subject of study can usually be counted or collected without too great a disruption of the associated environment. Sampling of plant root systems, however, is always difficult and laborious and often destructive (Head, 1971; Olsen, 1963). This study required the exposure of fine rootlets to a depth of about 10 cm. The biomass of the exposed root system had to be estimated and the plant parts containing insects removed. Such a task may have been possible with some form of water pump; however, the facilities and manpower were not available, especially at the frequency required. Sampling therefore was necessarily destructive. Sections of the root system were removed from the plant for study in the laboratory. The removal of a portion of the root system is not only a loss in itself but it also severs the necessary link between the tree and any distal portion of the root system to which it was joined. A small number of proximal samples could rapidly deplete the host tree and the insect population associated with it. This may be acceptable in a high density stand of a commercial crop; however, the study area was situated in a State forest with only a limited number of trees available. Wholesale destruction of the flora is prohibited and would have drastically reduced the suitability of the study area.

Root systems tend to be hidden to some extent in the soil. Being blind to what one is sampling is an effective way of diminishing sampling bias; however, it also relinquishes a frustrating number of 'empty' samples, especially at the outer limits of the roots' distribution. Added to this is the problem of identification. Root systems do not reflect the obvious morphological variation of aerial plant organs. The buffered soil environment does not induce gross morphological differences between plants; especially plants of the same genus. In the study area at least one species of Metrosideros (usually M.diffusa) was always closely associated with M.robusta (Plates 6 & 7). was often so close that only examination under a dissecting microscope could determine on which plant galls were present. Whilst larger roots of Metrosideros species were distinguishable from one another, no morphological differences were apparent between fine roots (on which galls were formed). The fleshy roots of tawa (Beilschmiedia tawa), the next most common associate species, were readily identifiable.

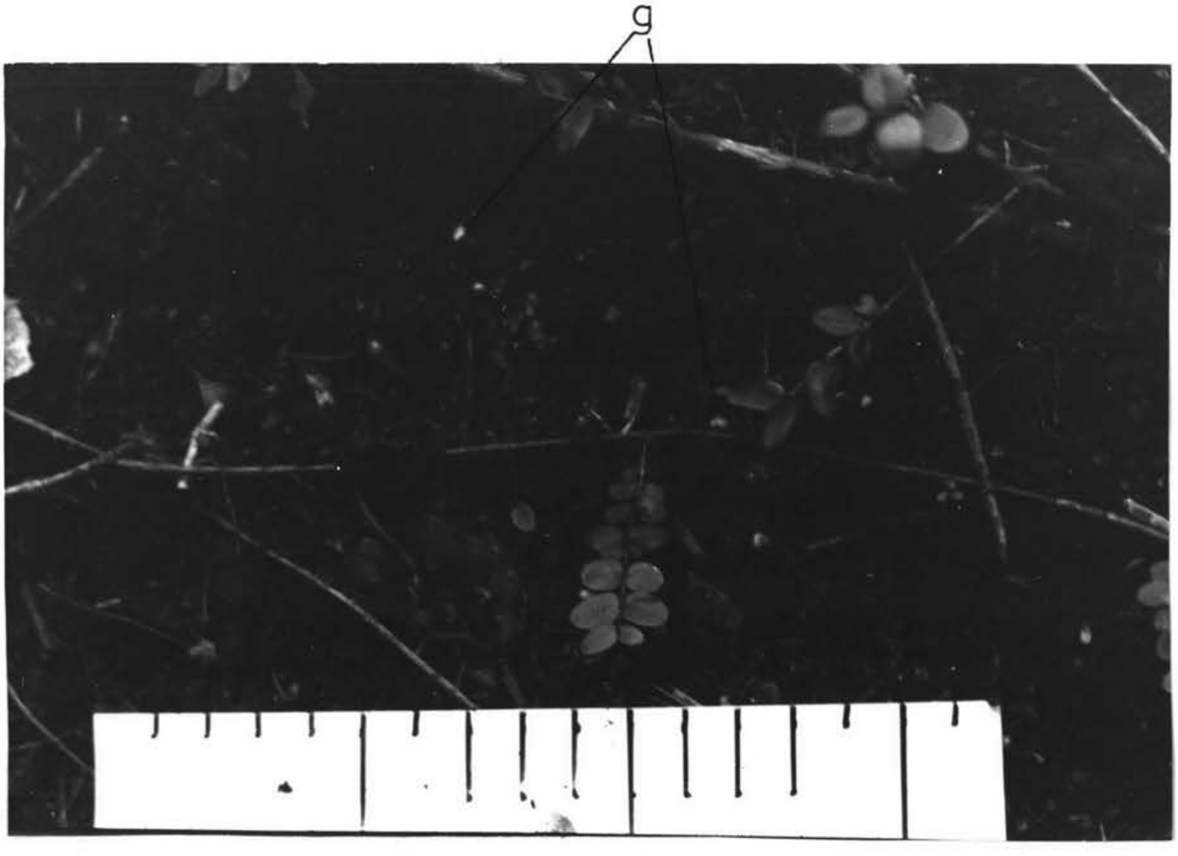
The small size of the insect was not necessarily a handicap to sampling but rather to the analysis of the samples taken. Galls ranging in size from 1 to 4 mm, and containing larvae 0.1-2 mm in length, had to be measured and dissected, and the larva removed, mounted and measured. The process took 2-3 days for 100 larval measurements. The gall-forming habit, however, does have some advantages for the observer. The insect is confined to one position for the greater part of its life cycle so that dispersal effects are minimised. The galls can be readily counted and density and, in some cases, mortality, can easily be measured. Many aspects of population dynamics can therefore be sampled in a single well-timed sampling effort. The measurement of a few 'key factors' (Morris, 1959), such as parasitism and dispersion, could provide the information for the prediction of population in the next generation.

The sampling programme of this study was required to yield information on the life cycle of P.radicis while simultaneously recording aspects of the population's dynamics

Materials and Methods

Initially the problems outlined above, especially root identification, precluded the collection of any truly quantitative data. Usual sampling methods record the change in numbers and development of the insect with respect to some host or abiotic parameter. The problem of host identity prevented a simple 'insect per unit host' approach, while the patchy

6



7

Plates 6,7 Two Metrosideros species commonly found in the study area.
6, M. perforata; 7, M. diffusa - illustrating the close association possible with the roots of M. robusta.
(g = galls)

distribution of the host's roots would require an impossible number of random soil samples to be analysed.

The following sampling programme was initiated. Two trees were chosen at random, from those on the north bank of the river (Figure 2) before each fortnightly (approximately) sampling expedition. Sections of gall-bearing roots from the trees were taken back to the laboratory. Only a small number of galls could be analysed, so to remove any clustering effects the galls were stripped from the roots, placed in a jar and covered with water. The galls were vigorously stirred while sub-samples (approximately 40 galls) were withdrawn with a small sieve (5 cm diam.). After each scoop the 'catch' was measured, dissected and various larval measurements taken (length, breadth, colour, head capsule width, spatula length, etc.). The process was continued until 100 live larvae had been encountered. About half-way through the study a process of identifying the host-plant material was developed. While the sequential sampling continued unchanged a quantitative sampling method was prepared for a single sampling effort.

Completely unbiased, unrestricted random sampling would be impractical and virtually useless in such a sparsely distributed population. The galls were obviously restricted to the root system of the host tree. The root density, on the other hand, decreases with the distance from the trunk. At the outer limits of root extension (up to 20 m, Section 1.3) the chance of encountering roots or galls would be slim. Completely random sampling on a grid basis would probably result in a majority of samples having very few galls, or none at all. A method of stratified random sampling was therefore developed. The tree was set in an imaginary grid of concentric circles, about the trunk, which were dissected by lines representing cardinal compass points (Figure 11). There were 10 concentric circles 1 m apart dissected by 10 lines 36° apart so that any two digits from a table of random numbers (Coulden, 1952) gave a

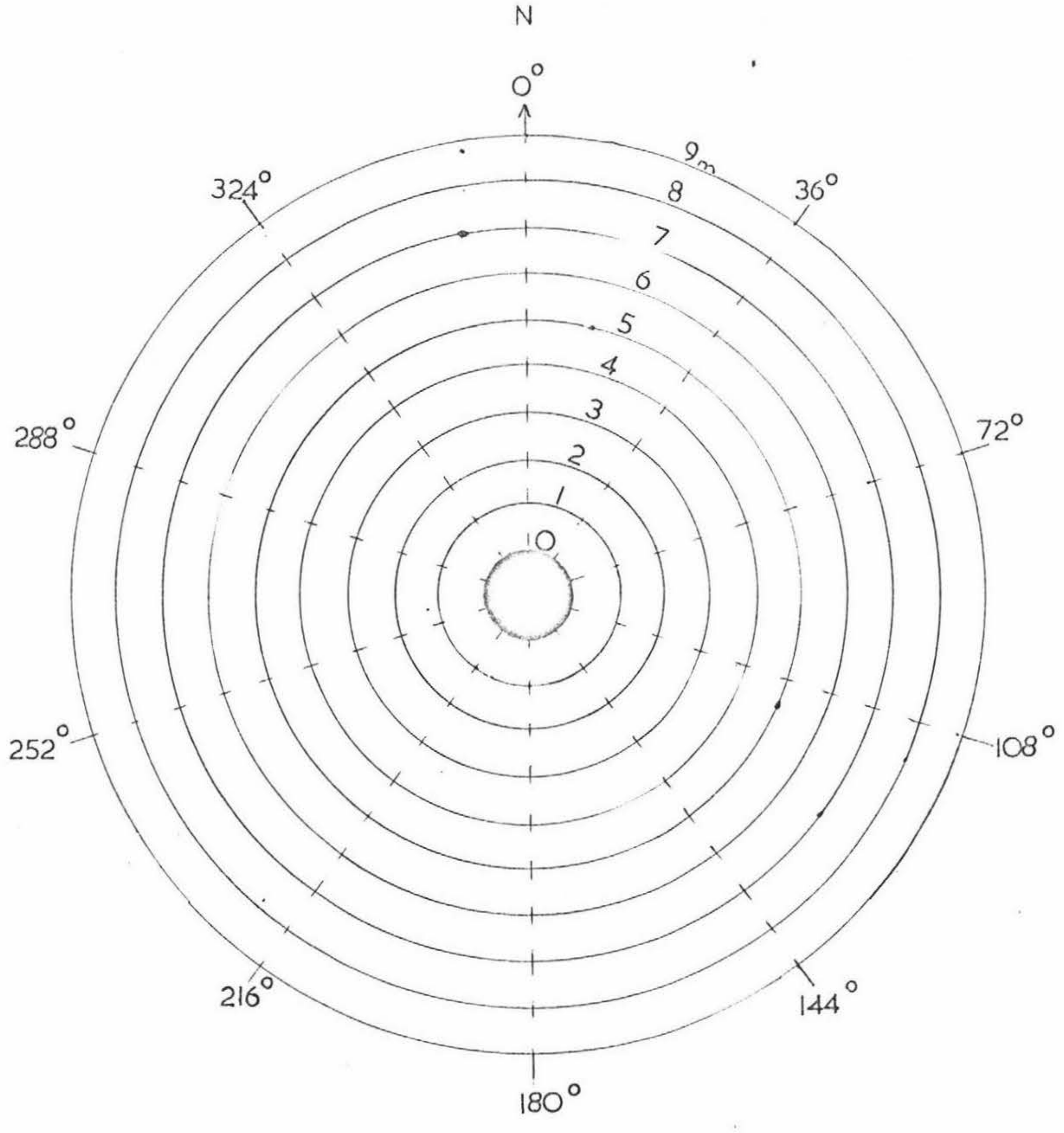


Fig 11 The sampling grid used during the analysis of root and gall dispersion about individual trees.

grid reference. The first digit represented the direction and the second the distance from the tree trunk. e.g. 05 represents a point 5 m from the trunk in a northerly direction, 15 represents a point 5 m at 36°N, 55 = 5 m south, etc. By this method sampling was weighted so that more samples were taken per unit volume nearer the tree's trunk.

Three trees were chosen at random from the north bank and 25 samples/tree were taken. Each sample consisted of three cores, from a 10 x 10 cm soil corer, arranged in a triangle with cores approximately 10 cm apart. The three cores were sieved through a 2 x 2 cm mesh, labelled, and packed in a plastic bag and brought back to the laboratory. A small soil sample (approximately 10 g) was taken from each sample in a small glass vial for moisture and ash analysis (Section II5). In the laboratory the roots were washed and the roots of Metrosideros sp. removed. Galls were removed from the roots, if present, and stored in alcohol. The roots of Metrosideros spp. less than 2 mm diam. were weighed and stored at -10°C until treated.

The treatment involved grinding each root sample with approximately 5 ml hot 70% ethanol (EOH, approximately 50°C) in a pestle and mortar. The mixture was transferred to a large tube (20 x 3 cm) and approximately 3 ml EOH/g of root added. The tube was loosely stoppered with a ground glass top and allowed to reflux for 48 hours at 108°C in an electric frypan filled with sand (Plate 8). Each batch of tubes (up to 20 could be accommodated at one time) was accompanied by similarly treated known weights of M.robusta root (1, 2, 3 g standards). These roots were chosen selectively in the field and could be traced as part of the root system sampled.

After refluxing the tubes were decanted and the supernatant evaporated to dryness at 108°C under reduced pressure (tap-vacuum pump). The residue was taken up in 5 ml EOH and 5 ul spotted on No. 3 Whatman chromatography paper. The chromatographs were run descendingly in two dimensions using n-butanol:acetic acid:water (6:1:2) and secondly 6% acetic acid (v/v).

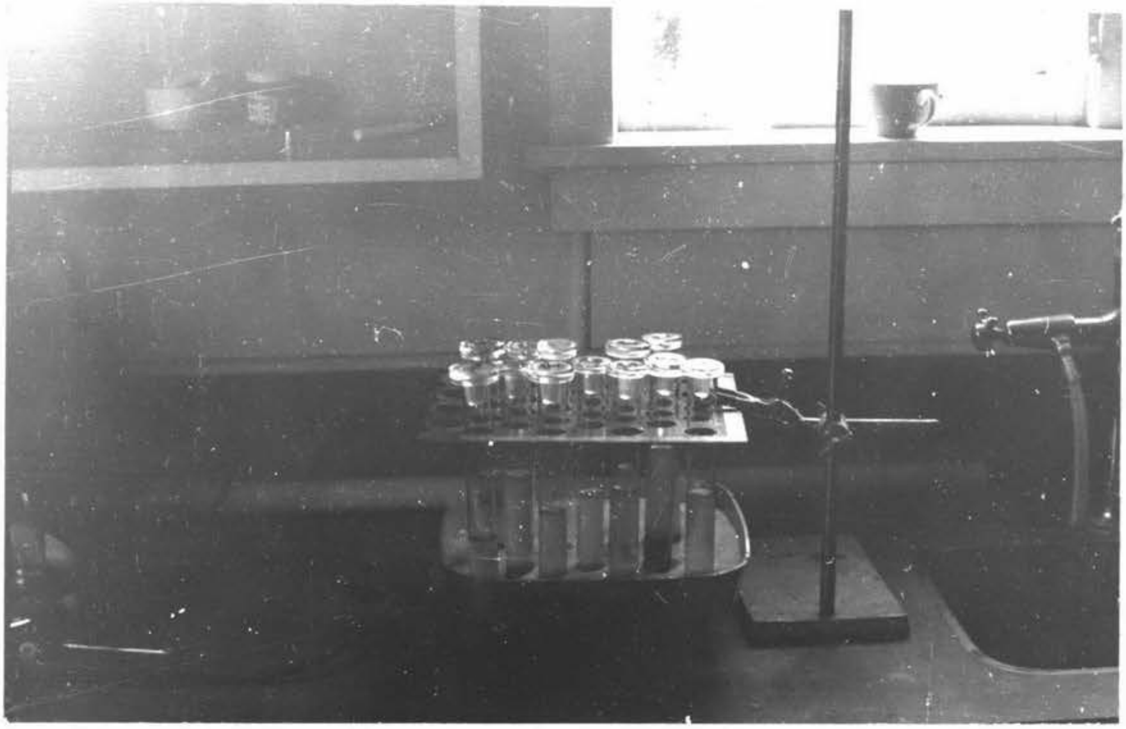


Plate 8 Apparatus for the simultaneous refluxing of a number of root samples.

The chromatographs were dried between runs and observed under an ultra-violet (u.v.) light.

The spot utilised for identification (Figure 12) was present only in *M.robusta*. It gave a bright pink fluorescence in u.v. which turned yellow under fuming ammonia (probably a free phenolic acid, Harborne, 1964). The strength of the spot was assessed by comparison with the standards and marked <1 , <2 , <3 + >3 g correspondingly. A semi-quantitative method following that of Keith *et al.* (1958) was tried in an effort to gain better sample definition; however, it proved too variable and time-consuming to apply to such a large number of samples.

Sampling of the forest insect fauna was achieved throughout the study by the use of two Malaise traps. The traps were basically tents (approximately 2 x 1 x 1 m) with two open sides and a hanging central panel (Plate 10). The traps were inclined with a collecting bottle containing alcohol at the apex. They were placed at positions M1 and M2 (Figure 2). M1 was in open forest on a small spur and M2 was in an open area below Tree 8.

Results

The results obtained using these sampling methods are described in the proceeding sections.

The results of the chromatographs of the phenolic compounds of the three *Metrosideros* species are illustrated in Figure 12.

Discussion

The sub-sampling of samples (used to elucidate the life cycle) proved to be reliable although it was slightly biased towards the collection of old galls (Appendix 2). These galls were empty and tended to float in a layer through which the sieve had to pass on withdrawing. The old galls,

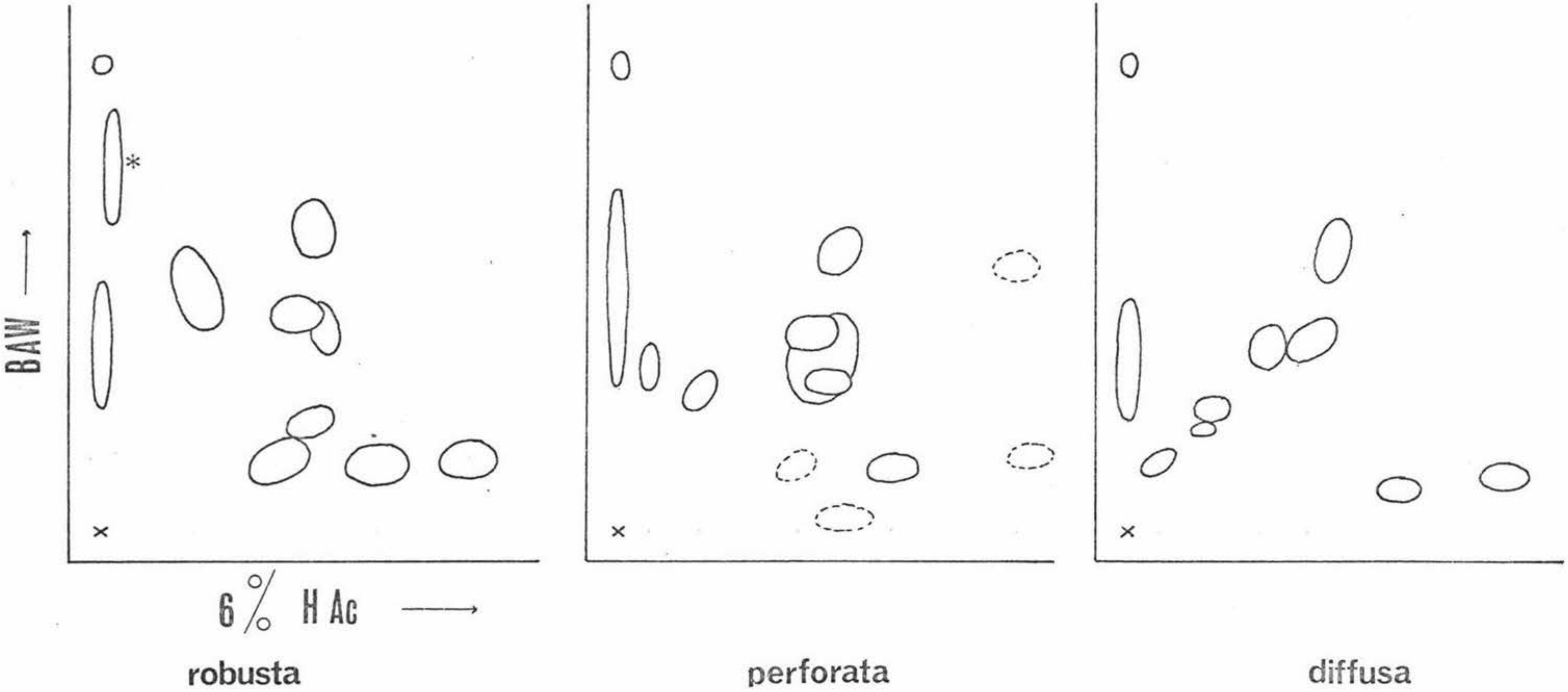


Fig. 12 Diagram of chromatographs showing the distribution of phenolics in 3 Metrosideros species.

* phenolic compound present only in M.robusta
B.A.W. = butanol, acetic acid, water (6,1,2)
6% H.Ac = 6% Acetic acid
x = initial application

Plate 10 A malaise trap (M1 Fig.) used during the study.

however, were of little relevance to the results required and the correction factor was not applied in later samples.

The method of grid sampling about a tree was satisfactory (Section II.5) but still resulted in a large number of samples containing neither galls nor roots. Similar methods have been used before to reduce sampling variance (Southwood, 1966). Prebble (1943) found that satisfactory estimates of sawfly pupae were obtained only if sampling was restricted to areas around the base of a tree.

The quantification of roots in samples appeared to be fairly effective within the limits imposed. The method, however, is too gross for good statistical correlations. In Section II (Dispersion) gross chromatography results are used in graphs and raw weight data is used in statistical analysis. This was not considered to be too unreasonable as the root systems of the two confusing species were rarely as extensive as that of M.robusta. Samples, especially large samples, undoubtedly contained more M.robusta than M.diffusa or M.perforata. Very small samples (less than 0.2 g), although producing no identifiable spot, were classed as <1 rather than 0 to indicate that they were present. Samples having categorically marginal spots were placed in the higher class.

It is interesting to note that a virtually identical method to the one used here was recently adopted to determine root density relationships between two species of Eucalyptus (Chilvers, 1972). Phenolic compounds are favoured as phenological markers because of their structural variability, widespread distribution, chemical stability and ready identification (Levin, 1971).

The Life Cycle of P.radicis

Life history studies of any organism provide a basis for the relevance of any future investigation. Zondag (in Wyatt, 1963) reported that each gall induced by P.radicis enclosed only one larva. The larva pupated within the gall and the majority of adults emerged in the second week of February. He also stated that galls collected in November showed evidence of a recent emergence..

In view of such sparse information and the breadth of the present study, it was considered necessary to gain as much information as possible about the insect's life history.

Materials and Methods

Data about the life cycle were obtained from the sequential sampling programme outlined in Part 1 of the preceding section. In the laboratory galls were measured, dissected under a dissecting microscope, and then the larvae or pupae were removed and measured under a compound microscope. As this sampling technique did not detect eggs and rarely revealed first instar larvae, the presence or absence of the first larval instar was assessed by examining selected root samples under the dissecting microscope. Towards the end of the study (in conjunction with the investigation of nematode parasites) it was found that first instar larvae were readily extracted from a soil and root sample by using a Baermann funnel.

Eggs could not be detected in the field and observations were made from laboratory-induced ovipositions (Section II.4)

Adult emergence was forecast from the analysis of field studies and emergences in laboratory rearing cages (Appendix 6). It was confirmed in the field with catches from hand nets and aspirators. Two Malaise traps were also in use. Unless otherwise stated, the duration of individual stages was taken as the time from the first appearance of the stage to the first appearance of the proceeding stage.

Results

The results of the fortnightly samples showing the development of the population are collated in Figure 13 (Data Appendix 7). The fortnightly samples over winter have been averaged, while the summer samples have been left to show pupal development more clearly.

Eggs of P. radicis were only detected in the laboratory. First instar larvae, however, were found on root material, at the initiation of a gall, or they could be extracted from a soil and root sample with a Baermann funnel (Section II.9). The larvae were situated on developing rootlets just behind the meristem. The root grows in a spiral around the larva. During the second larval instar the root continues to grow in a spiral, expanding diametrically, to eventually enclose the larva. Gall growth is completed in the third larval instar and finally resembles a smooth lens-shaped structure 3-4 mm in diameter.

The results of gall and larval measurements (Figure 14) show that there is a significant correlation ($r = 0.696$) between gall size and the development of the larva (i.e., a larger gall contains a larger-older larva). Galls containing first and second instar larvae are creamy white; however, the gall gradually darkens to a brown colour during the third instar.

Galls containing pupae showed a strong binodal size distribution (Figure 15). The sex of the enclosed pupae showed that this was a significant ($t = 10.365$, 19 d.f., Table VIII) sexually induced dimorphism. Larger galls contained female pupae.

Towards the end of the third larval instar an aperture, later to be used as an exit hole, is cut in the 'edge' of the gall. It remains covered by a thin layer of undisturbed plant tissue until adult emergence. After the exit hole is cut the larva assumes a C shape (ventral surface

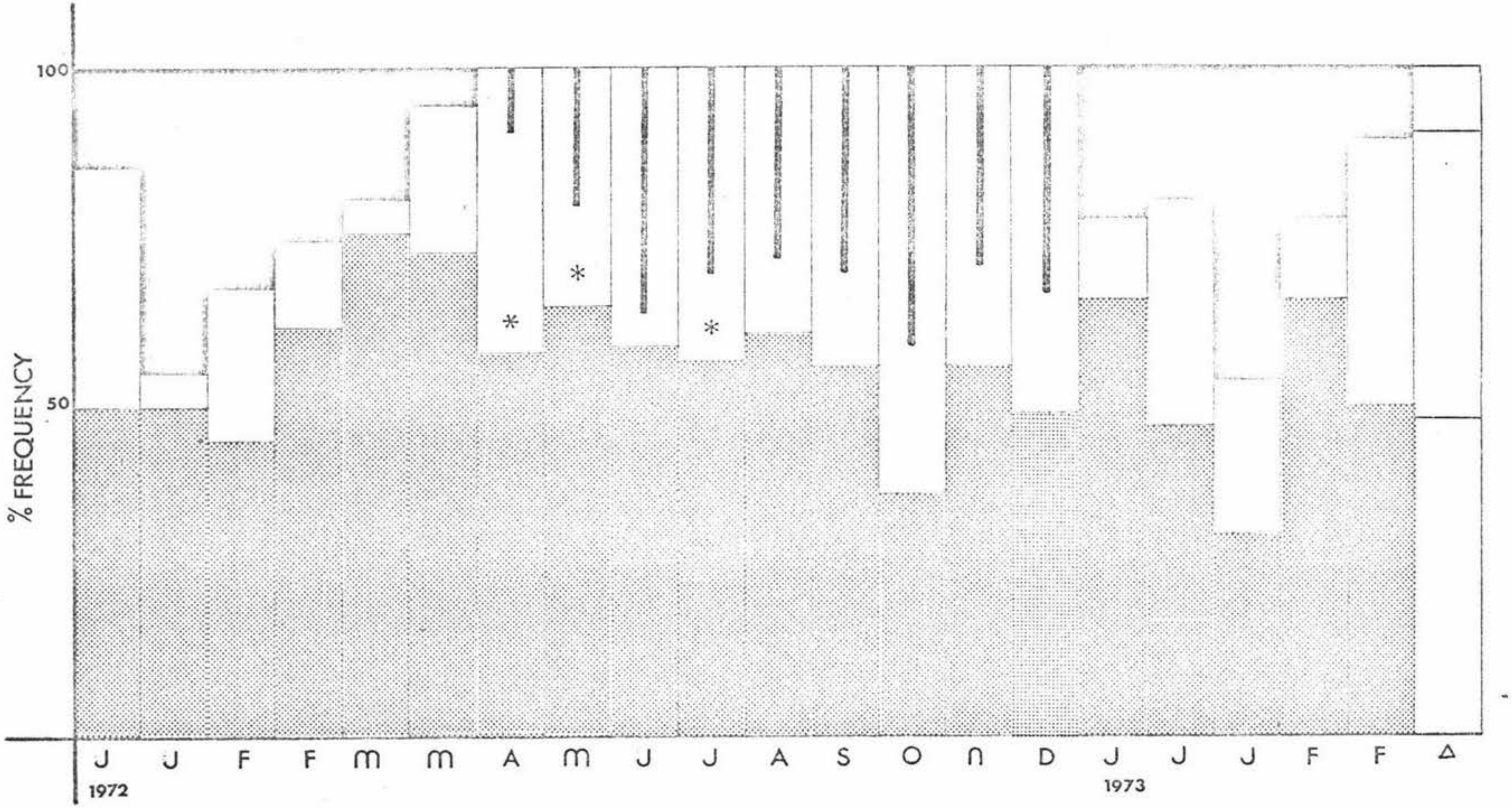


Fig.13 The change in the population structure of P.radicis with time.
* = first instar larvae ⁙ = second instar larvae ☐ = third instar larvae T = exit-hole cut ■ = pupae
• Δ = results of dispersion sample 13.2.73 (Section II.5)

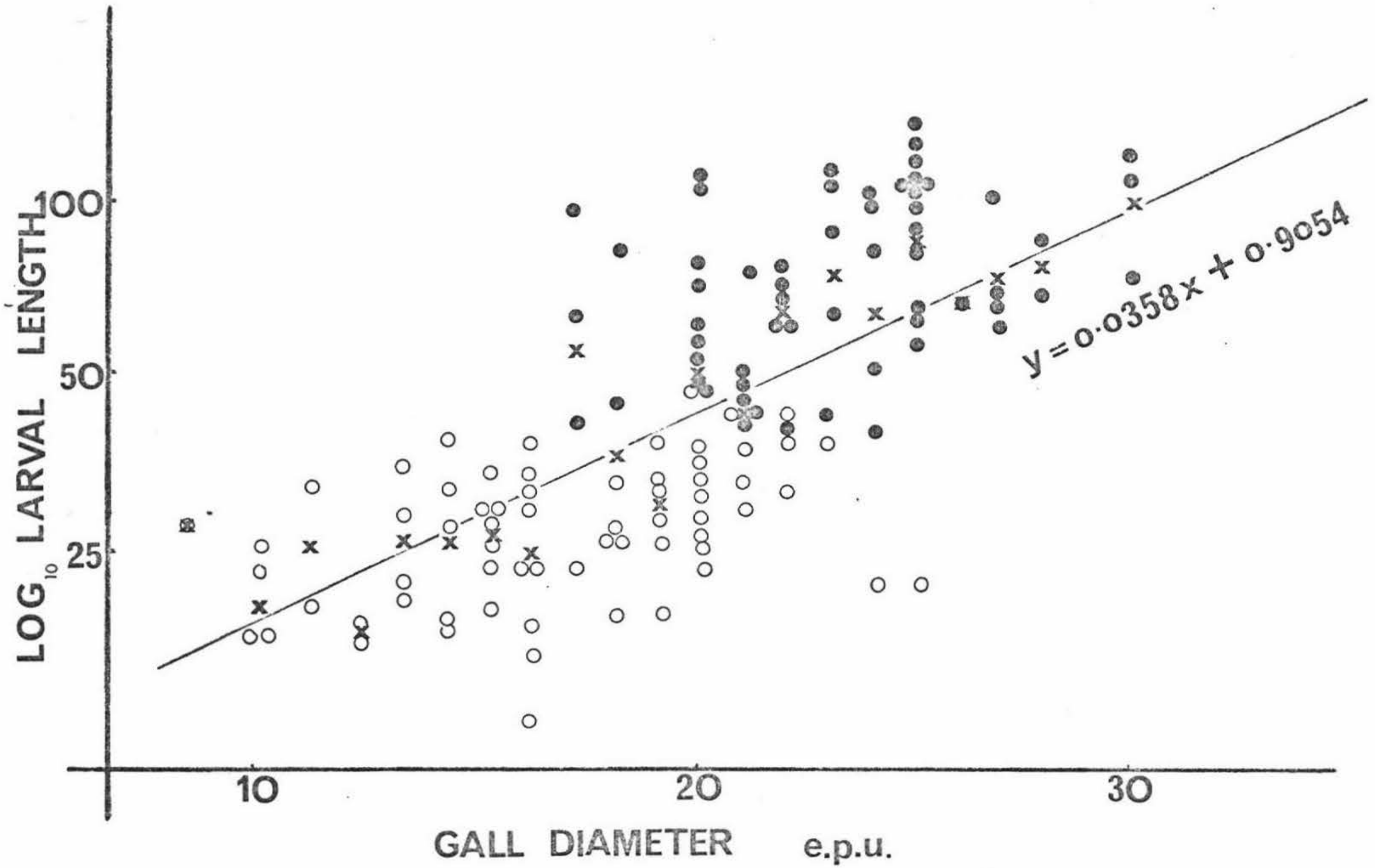


Fig. 14 The correlation between larval length and gall diameter.
O = galls of second instar larvae ● - galls of third instar larvae
X = mean larval length for a given gall diameter
(n = 130, r = 0.696 , st. err. = 0.186)

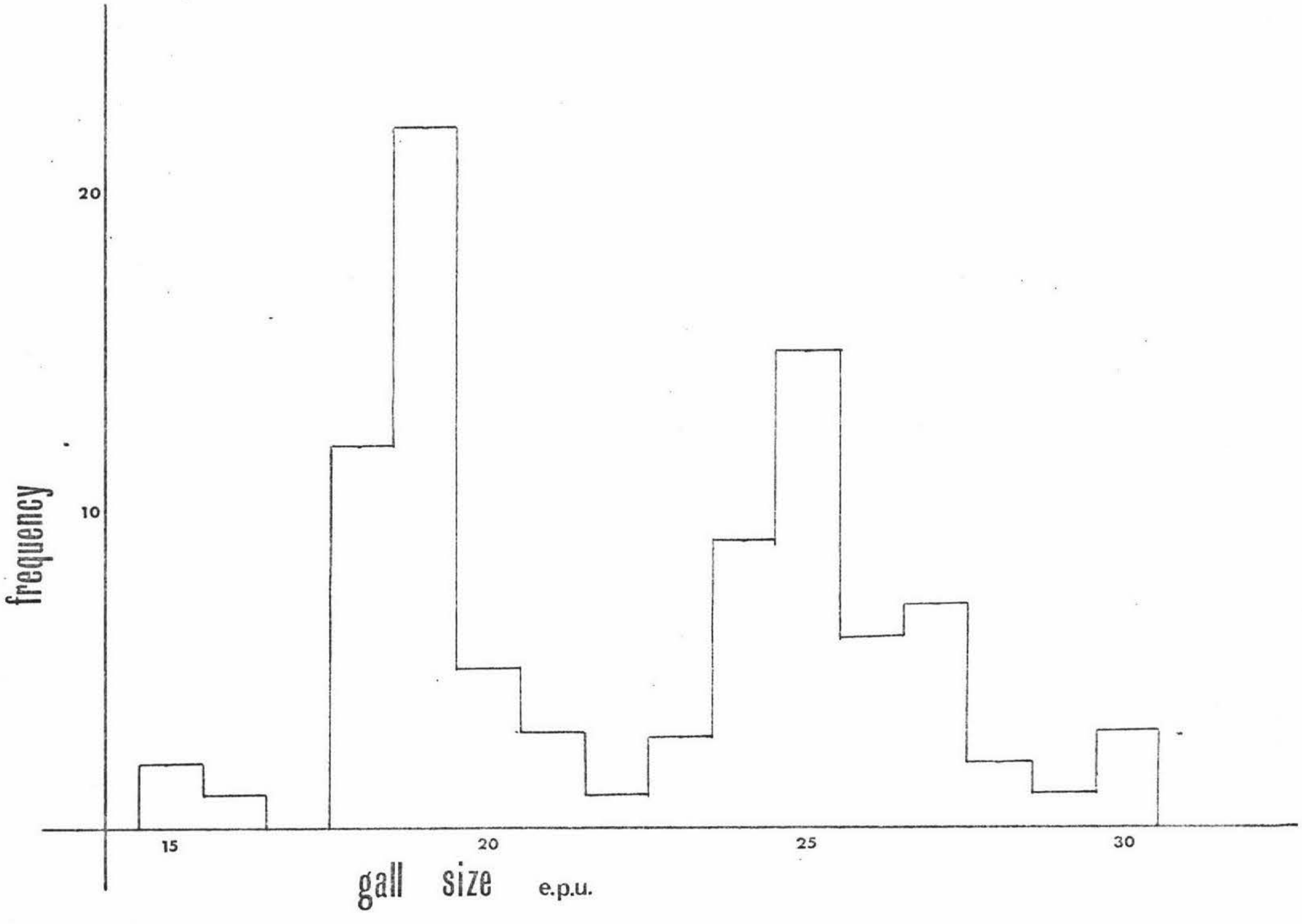


Fig. 15 The size distribution (diam.) in a sample of mature galls.

GALL SIZE (mm)	
containing female pupae	containing male pupae
3.50	2.90
3.85	2.10
4.35	2.70
3.00	2.35
3.90	2.68
3.52	2.15
3.71	1.90
3.45	2.80
3.60	2.40
3.45	2.25
4.00	2.65
3.42	2.65
3.16	2.80
3.63	2.75
3.54	2.90
3.78	3.10
3.80	2.90
3.18	2.75
3.64	2.72
3.80	2.90
$\bar{x}$ 3.61 ±0.06	2.59 ±0.06
$\bar{x}_1$ = 7228	$\bar{x}_2$ = 5235
t = 10.365	

TABLE VIII GALL SIZE WITH RESPECT TO THE SEX OF THE ENCLOSED PUPAE OF P.radicis outermost) as far removed from the exit as possible. Prior to pupation the larva straightens, becomes distended, and pales anteriorly. During this prepupal stage the dorsal ocular stigma divides and the two halves migrate ventrally to lie near the base of the sternal spatula. Pupation takes place within the gall with the anterior region of the pupa close to the exit hole. Although the cuticle of the head and thorax is sclerotised the abdomen remains soft and is capable of flexing. The anterior edge of the abdominal segments bears a number of small papillae which probably aid movement within the gall. Prior to adult emergence the thin operculum of plant tissue covering the exit hole is broken and the pupa protrudes through the aperture. The adult emerges from a longitudinal fracture of the thoracid dorsum. Pupae removed from galls and placed on damp filter paper experienced difficulty during ecdysis and adults usually emerged with damaged or deformed appendages.

The duration of the larval instars is discussed below. The prepupal period is relatively short (1-2 weeks) as pupae were recorded with the first samples containing prepupae. The pupal instar has a duration of about four weeks. Adult flight and oviposition extends over five weeks as measured from the peak of pupal frequency to the disappearance of pupae. The individual life span of adults is extremely short (1-3 days, Section II.4) No eggs were found in the field; however, in the laboratory eggs showed signs of hatching after one week (Section II 4).

Discussion

It can be seen that *P.radicis* has only one generation per year, with the adults emerging from mid-February to mid-March. The emergence occurs during the high February temperatures and is followed by the moister conditions of March (Figure 3). The evidence for an earlier November emergence, suspected by Zondag (in Wyatt, 1963), probably came from the presence of old galls. These dark-brown empty galls usually predominate in any sample taken. During this sampling programme old galls outnumbered fresh galls by about 3 to 1, indicating that either old galls have a fairly slow degradation rate (i.e. three years if the population is stable) or the previous year's population was exceptionally large.

The detection of first instar larvae was fairly spasmodic, but it was noted that this stage may be prolonged and that a percentage of the population exists in this stadium during the winter months. Essentially the first instar larvae represent a free living stage with reasonable powers of locomotion (Section I.4). From the observations made it was concluded that the first larval instar is responsible for the initiation of the gall. The stimulus for gall production must be retained until late in the third instar. That there is a sexually induced dimorphism in gall size indicates a sexual differentiation in the larvae, i.e. presumably some time before gonadal development. No marked larval dimorphism was detected (Section I.4). The correlation between larval sex and development and gall size and colour was used in later experiments (Section II.4)

The exit hole created late in the third instar is probably cut with the sternal spatula. This is a ventral bilateral cuticular structure which develops in the third instar and is fully sclerotised in mature larvae (Section I.4). The function of the spatula has been variously interpreted as an organ of escape or locomotion (Imms, 1967). A plot of tine length against larval length (presumed equivalent to age) shows that tine length decreases with age (Figure 16). Similar tine wear has been observed in Dasineura tetensi (Pitcher, 1957) and it is generally conceded that the spatula abrades plant tissues to aid the escape of the midge from the gall (Anderson, 1935; Hedlin, 1961). From the results of the removal of P.radicis pupae from galls it would appear that the constraint of the gall facilitates eclosion.

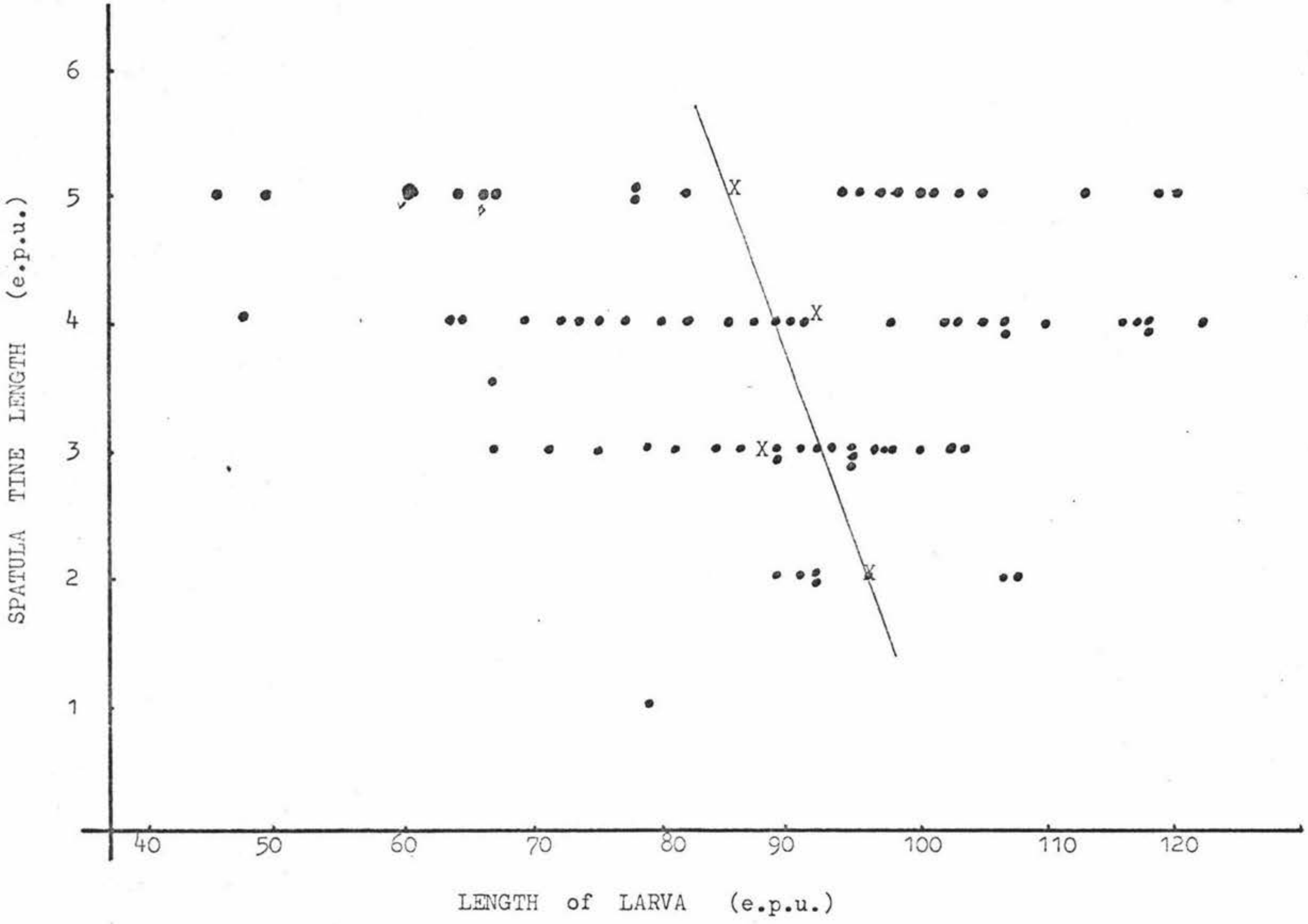


Fig. 16 The relationship between sternal spatula tine length and larval length (age).
(X = average)

The duration of individual stages in the life cycle was difficult to assess accurately. The character of the samples remained remarkably constant, approximately half being second and half being third instar larvae, thoughout the year. This suggests a slow continuous recruitment from one larval stadium to the next, or a continuous mortality in one or more of the larval stages. Both processes were probably operating; the prolonged first larval instar supplying new recruits for the second instar which in turn develop to the third instar, with each experiencing some mortality. The first larval instar was found from April to July and may be virtually quiescent (i.e. facultative diapause) during the winter months. Those larvae enclosed within galls may be better protected from extremes of weather and may continue to develop. There appears to be a slow recruitment into the third instar during this period, climaxing in October when temperatures and root metabolism rises (Figures 3 & 32). As temperatures continue to rise, quiescent first instar larvae may become more active and the numbers of the second instar larvae increase to achieve parity with the frequency of the third instar in November.

The incidence of diapause in the New Zealand insect fauna is comparatively low (Dumbleton, 1967). A small proportion of the P.radicis population, however, appears to enter diapause as third instar larvae. The proportion (ca. 10%) is taken as the percentage of the third larval instar in the population which, at the end of adult emergence, remain in galls in which exit holes have been cut (Figure 13). The proportion is fairly low when compared with diapause in northern hemisphere cecids (Table IX). The advantages of diapause to such frail insects, which require a close phenological synchrony with a host, is obvious and its presence in the P.radicis population should provide adequate insurance against an untimely emergence.

Adult Longevity, Mating and Fecundity

Behavioural observations of adult cecids are rare as the adults are frail and short-lived. However, a knowledge of an insect's fecundity, longevity, and mating habits is necessary for the construction of census budgets. These aspects of the biology of P. radicis are investigated in this section.

Materials and Methods

All observations, unless otherwise stated, took place in the laboratory. Eggs, mating and oviposition were not observed in the field. Adult insects used in experiments were captured with an aspirator after emerging from galls housed in rearing cages (25 x 25 x 40 cm - two sides and top panelled with glass). The galls were collected from the study area approximately two weeks prior to adult emergence.

Longevity

Groups of 4 or 5 males and females were collected in the morning and placed (sexes separate) in two small (5 cm diam.) plastic tubs. Each tub had a circle of moist filter paper stuck inside the lid and a small plastic container (2 x 2 cm) filled with 10% sucrose solution. Four small holes in the sides of the tub allowed ventilation. Adults placed within the tubs were observed each morning and afternoon and the time at which they died was noted.

Mating

Tubs similar to those above, but without filter paper or sucrose, were used to observe matings between adults of P. radicis. Female(s) were introduced into the tubs, allowed to settle and then followed by male(s). Three observations involved single couples in separate dishes and two involved single males with two females. Two females in single pairings and one female in a double pairing, were damaged. Sex ratios were collated from the three rearing cages of the 1973 flight season.

A sex attraction experiment was done in the field during the 1973 flight period. A number of large brown galls thought to contain pupae (Section II 3) were mixed with damp vermiculite and placed in small plastic petri dishes. The bottoms of the dishes (5 cm diam.) had been removed and replaced with cotton mesh ($10/cm^2$). Control dishes were identical but contained damp vermiculite and pieces of *M.robusta* root and old galls. Each dish was mounted on a white hardboard square (12 x 12 cm) which was greased with a film of Atlantic No. 1 chassis grease. A treated and a control trap were suspended about 10 cm above the ground and about 30 cm apart on a wooden T (Plate 11). Four traps and their controls were placed in the vicinity of galled roots about Tree 1. Insects alighting or colliding with the greasy surface were trapped. The traps were recovered after 11 days and washed in petrol to dissolve the grease. Adults of *P.radicis* were then counted.

Fecundity

Total potential fecundity was recorded by dissecting newly emerged or mated females in a drop of insect saline and counting the eggs from the ovaries.

Live mated females were placed in petri dishes provided with moist filter paper to observe oviposition.

In an attempt to determine the natural site of oviposition (and at the same time infest cuttings of rata), mated females or males and females were caged with four cuttings of *M.robusta* in large paper cups (15 x 12.5 cm). Cutting propagation is described in Section . The roots of the cuttings were exposed and standing on moist vermiculate and each cup was covered with a small plastic bag to confine the midges. The adults were left in the cages for about 24 hours. The roots of the cuttings were then inspected for eggs. The plants were either (3 plants) returned to the glasshouse and potted in a loose mixture of peat and vermiculite, or retained (1 plant) in the laboratory for further observations on egg development. The observations entailed a daily viewing under a dissecting microscope with the plant remaining undisturbed, apart from watering, in its original cup. During the 1973 flight season three cuttings were potted in wet vermiculite and placed about Tree 1.

Results

The results of the studies of P.radicis are collated in tables X-XIII and can be compared with the results of studies of other cecids in Table IX.

In the rearing cages males appeared 1-2 days before females (Appendix 6). Males, however, as in the field, are more conspicuous in that they fly above the litter or rest on the sides of the cages. Females stay within the litter, sometimes at considerable depth, and have to be 'dug' out before capture.

Longevity

The results of the longevity studies of adult P.radicis are shown in Table X. Females were slightly longer-lived than males (average 2.5 and 1.5 days respectively).

Mating

A number of apparently successful matings were witnessed. During mating the males mounted the females from the rear and the male genitalia normally carried dorsally were turned ventrally to clasp the genitalia of the female. Males which could not mate successfully with damaged females moved in an agitated manner around the female clasping wings, head, etc. with their genitalia. Unmated females continually extended and withdrew their posterior abdominal segments. A male in the presence of two females paired with both in quick succession. The females did not appear to be attractive to the males after mating had occurred. Generally

Species	Longevity (days)	Fecundity	Diapause %	Reference
Contarinia pseudotsuga	o 1-2 ♀ 2-4	95-121	present	
C.oregonensis	o 2.9 ♀ 3.7	119-396	52%	Hedlin, 1961
C.washingtonensis	o 1-9 ♀ 2-13	94-140	50%	Hedlin & Johnson, 1963
C.nasturtii	approx. 1	95 ±3	present	Readshaw, 1965
Dasineura sp.	1-1.5		present	Giese & Johnson, 1959

TABLE IX LONGEVITY, FECUNDITY AND DIAPAUSE RECORDED FROM OTHER CECIDOMYIIDAE

Sex	Longevity (days)	Range	No.of recordings
Male	1.5	0.5-2.0	10
Female	2.5	1.0-3.5	9

TABLE X   P.radicis ADULT LONGEVITY

Cage	No. ♂	No. ♀	Ratio ♂/♀
A	13	21	0.62
B	38	36	1.05
C	28	31	0.90
	79	88	0.89

TABLE XI   THE SEX RATIO OF P.radicis
AS RECORDED FROM REARING CAGES

Trap no.	'Baited' trap		♀ trap	Control trap		♀ trap
	♂	♀		♂	♀	
1	40	2	1	6	5	0
2	33	1	1	3	2	0
3	13	6	6	4	3	0
4	31	2	3	5	3	0

TABLE XII   RESULTS OF SEX ATTRACTION TRAP

♀	Egg count	Av. ±SE
1	124	
2	103	
3	110	116 ±4
4	125	
5	117	

TABLE XIII   FECUNDITY - RECORDED FROM DISSECTIONS OF FEMALE P.radicis

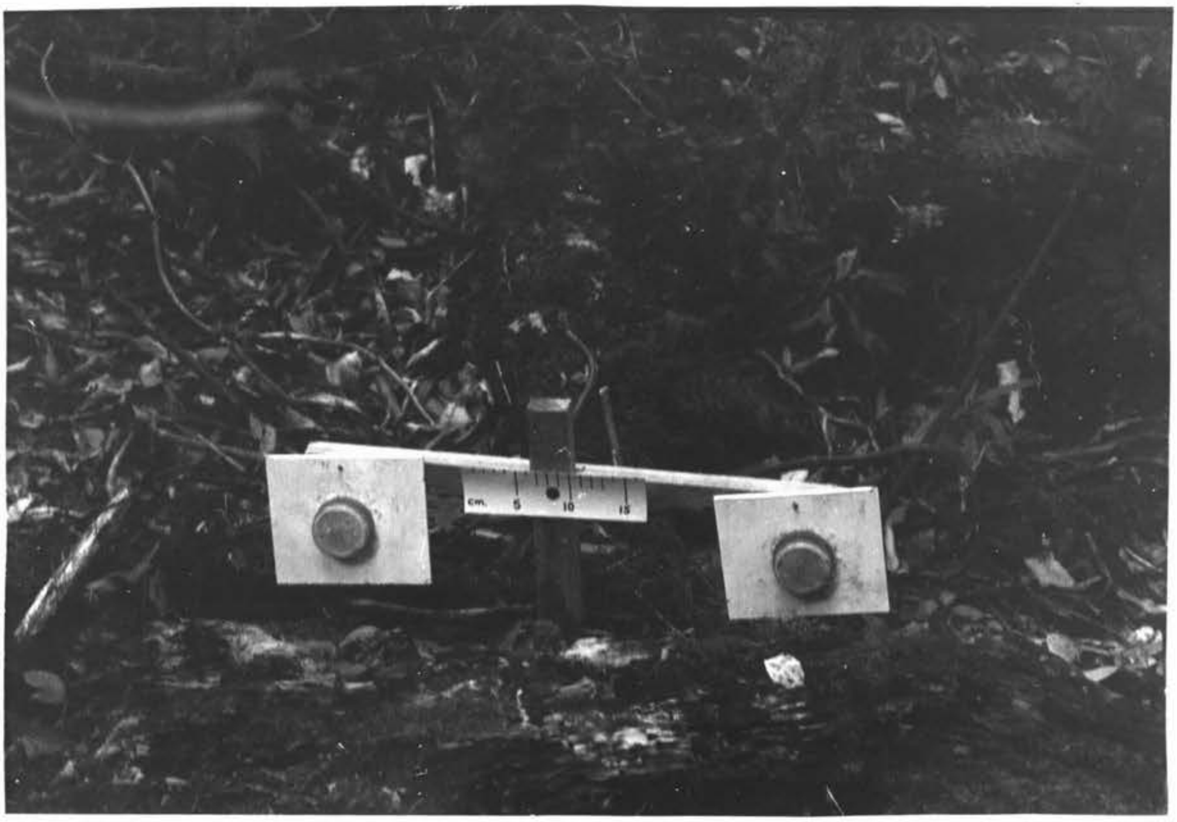
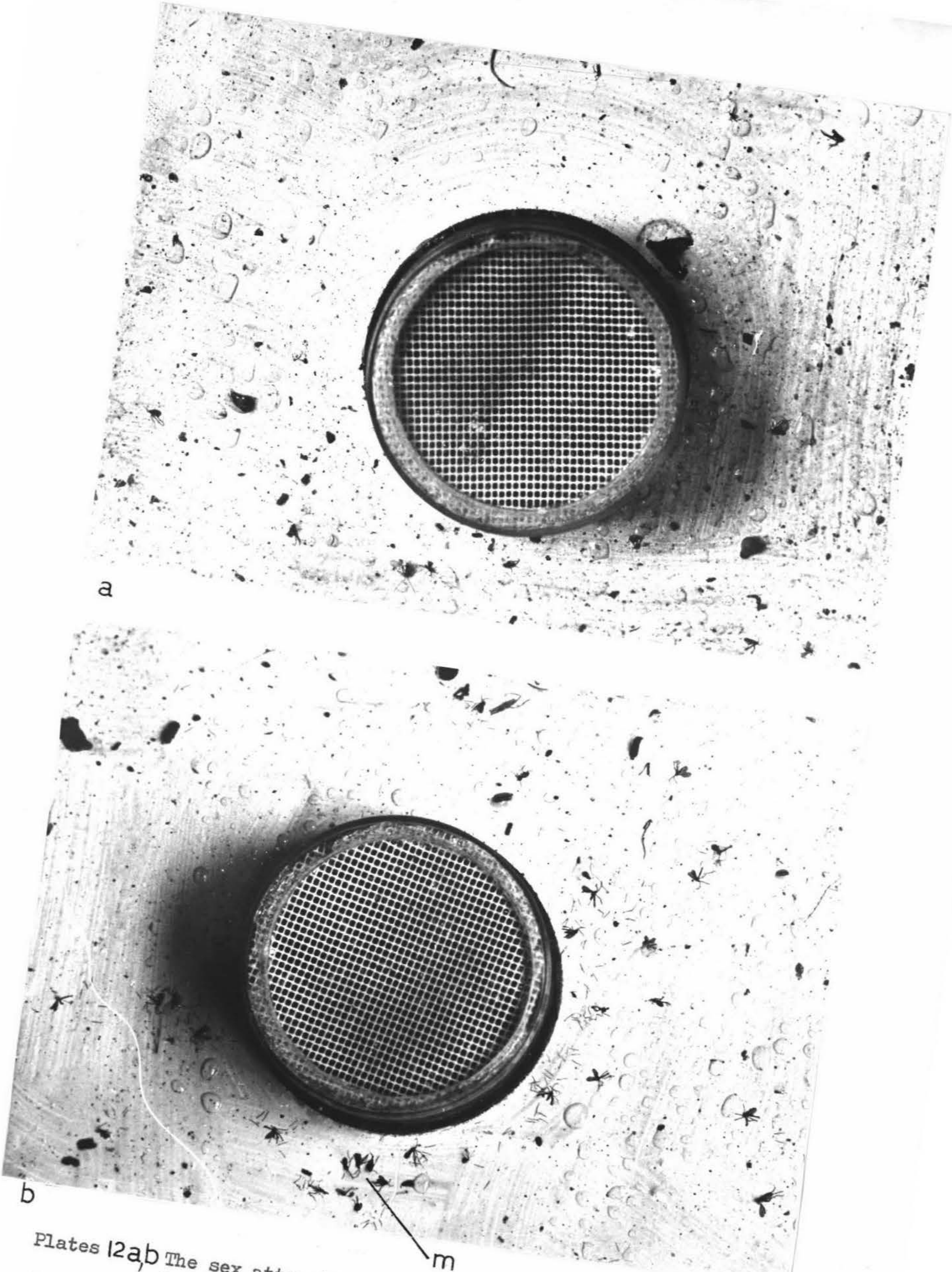


Plate 11 Sex attraction traps (baited & control) in the field about Tree 1.



Plates 12a,b The sex attraction traps ; a, a control trap
b, a baited trap showing a number of captured males (m).

matings were rapid and completed in 1-2 minutes.

The results of the sex attraction traps placed in the field are recorded in Table XI. A Students' t-test shows a significant difference ($t = 10.9$ 3 d.f.) between the numbers of males caught on traps containing females and control traps. (There was no significant difference between the number of females caught on both.)

Fecundity

The total potential fecundity of the five females dissected is shown in Table XIII. Average fecundity was 116 eggs.

Natality (the number of eggs laid by females) on filter paper varied. One female laid approximately 50 eggs in groups of 16-18. The average interval between egg layings was 32 seconds (averaged over 14 timings). Another laid a cluster of 27 eggs on the side of a petri dish above the moist filter paper. Females caged over rata cuttings laid eggs in clumps, in pairs or singly, on or near roots. Two readily observable eggs were laid at the base of a developing rootlet and were examined each day for 7 days. After this period the ocular stigma were visible through the chorion of the egg but the larvae had not hatched. None of the eggs laid on the roots or transferred to the roots from petri dishes survived to induce gall formation on the cuttings. Cuttings placed in the field during the 1973 flight period were not infested and at no time were eggs, mating or oviposition observed in the field.

Discussion

The fragility of the adults proved to be a major problem in these studies. Adults in rearing cages became trapped in the condensation on the glass panels and others, when caught with an aspirator, invariably lost legs or antennae. Legs were frequently lost when specimens were placed on moist filter paper. The adults had a very short life span and were very susceptible to low temperatures. Any attempt to maintain sufficient numbers of adults for a particular activity by a slight cooling invariably resulted in their death. Pupae were more resistant to cooling and could be kept up to a week at about 4°C without too great a loss. Pupae excised from galls, however, emerged with deformed wings and missing legs.

P.radicis adults represent a short-lived, non-feeding stage in the life cycle. As is commonly found with cecids (Hedlin, 1964; Hedlin & Johnston, 1963) the females are longer lived than males.

The males and females of P.radicis show a distinctly different pattern of behaviour. The males are found in sheltered pockets in the undergrowth above the soil litter layer whereas the females are found within the litter layer, sometimes at considerable depth. This cryptic habit may have resulted in some females being overlooked and biasing observations of emergence and sex ratio in favour of the males. The sex ratio from rearing cages was close to unity (0.89 male:female), with only slightly more females than males being captured.

From the observations of longevity, emergence and sex ratio, considered in the light of gametic efficiency, one would expect that males being shorter-lived, emerging earlier and yet being in approximately equal numbers to females, would mate more than once. This was borne out in the few observations of matings in the laboratory. The matings, however, occurred in an artificial environment and in three matings the females had lost legs and/or antennae.

The excited and inappropriate clasping of damaged females, and the complex array of sensillia on the antennae of male P.radicis, indicated the use of chemical cues in mating, or mate location. Male Cecidomyiidae are renowned for their elaborately looped sensillia (circumfila). The P.radicis male has three whorls of looped circumfila on each antennal segment (Figure 10). Each whorl consists of 9-12 loops. The antennae of the female are shorter and the two circumfila per segment are greatly reduced (Figure 10). The results of the field experiment indicate that some form of attraction is utilised by the male in locating the female. Unfortunately the nature of the attraction could not be followed up in the laboratory. Chemical attraction, however, must be strongly suspected. Firstly, because of the elaborate male sensoria. Secondly, males attempted matings with damaged, virtually immobile, females in the laboratory; and thirdly, visual acuity of the males appeared to be poor, with males attempting to clasp wings, head, etc. of damaged females. Within the traps, packed with damp vermiculate, auditory and visual cues would be difficult to effect. Considering the females' normal cryptic habit, chemical attraction would appear to be the only suitable means for mate location. Cartwright (1922) advanced some evidence for chemical attraction in the Hessian fly (Mayetiola destructor). He stated that males flew upwind, from up to 15 cm distance, to field cages containing live or dead females.

Total potential fecundity of the five females dissected is consistent with observations of other cecids (Table IX). Natality (the number of eggs laid) and fertility (the number of viable eggs laid) were more difficult to estimate. Natality observed in the laboratory was considerably lower than fecundity. However, the observations were recorded from damaged females ovipositing on damp filter paper.

The ovipositor of the female is quite extensible but it is soft and pliable and is unlikely to be capable of piercing plant or other solid material. The ability of the female to move within the litter to some depth must account, in part, for the depth at which galls are found (up to 10 cm, Section II.5)

In summary it is probable that the typical life cycle of P.radicis has a duration of one year. Eggs laid by early emerging adults probably hatch in one to two weeks and the first larval instar, present during the active root growth of the host, readily secure a feeding site. They may develop to the second instar or possibly the third instar before cooler temperatures limit root growth. With the warm spring temperatures and the resumption of root growth development is completed in time for the late summer emergence. The development of eggs and larvae from late adult emergences may be checked by the cool winter temperatures and slow root growth. The resumption of root growth in spring would stimulate larval development but some would probably be too late for that year's emergence.

The postulated spring generation of Zondag (Wyatt, 1963) would readily explain the observed overlap of larval stages. However, no further evidence of a bivoltine habit was discovered. A pupation period totalling six to seven weeks could hardly have been missed by the sampling programme and pupal paedogenesis was not observed. From Zondag's description the dark brown empty galls were undoubtedly old galls from previous generations. The smaller white galls of the second instar were not mentioned and may have been overlooked.

The Distribution and Dispersion of *P.radicis*

Populations at the outer limits of their species range are often exposed to 'disruptive' rather than 'regulatory' processes (Milne, 1957). In its optimal environment a species tends to be regulated by density-dependent factors such as competition, parasitism and predation. However, as the species moves from its optimal environment, disruptive factors, usually climatic extremes, tend to limit the population. Weather is usually considered the main disruptive process in that it has a relatively density-independent nature. A population at the limits of its range is uaually at the limit of its climatic tolerance and a relatively small fluctuation in weather conditions may greatly affect the population.

P.radicis has been recorded from a large area of the central North Island (Wyatt, 1963). The study area was situated in the south of the North Island so the distribution of *P.radicis* and the position of the study area with respect to the range of *P.radicis* was investigated.

Within the study area an attempt was made to relate the dispersion of *P.radicis* to some biological and physical attribute of the environment. Dispersion, or the pattern of animal distribution in space, is of considerable ecological significance (Southwood, 1966). It can be used to elucidate associations between individuals and their environment and can be correlated to events in the dynamics of a population. The initial dispersion of a population can be indicative of optimal conditions preferred by the insect, while a change in dispersion may reflect the effects of competition, predation, parasitism, etc.

Materials and Methods

Observations of the presence of galls on the roots of *M.robusta* were made in the Orongorongo Valley, Akatarawa Ranges, Tararua Ranges, Ruahine Ranges, Mamaku Ranges, Tokoroa and Coromandel Ranges (Figure 17).

A sample was also received from Bushy Park, Wanganui.

Within the study area dispersion was investigated with respect to macro- and micro-environmental parameters. The presence of P.radicis ('infestation') on individual trees in the study area was related to the age and health of the tree, and to the altitude, aspect, and soil moisture of the tree's site. Dispersion about a particular tree was related to the root density, soil moisture and soil organic content.

To assess infestation accurately a large number of samples would have to be taken per tree. With 20 trees to assess the time and effort required to analyse soil samples would be prohibitive. Infestation was therefore determined semi-objectively. For any tree the number of large (ca. 15 cm diam.) roots radiating from the trunk was determined. Observations were made on each root at 1 m intervals until 10 readings per tree were taken, e.g. for a tree with 5 major roots each root is sampled at 2 points 1 m apart, the first sample for each root being taken 1 m from the trunk. A tree with only one root would be sampled at 10 points along that root. The 'sample' consisted of an examination of about 0.5 m^2 at the sample point, for the presence or absence of galls. Infestation was ranked from 0 to 10: if galls were present at all points sampled a tree would get an infestation ranking of 10; if galls were present at nine points it would rank 9, etc.

The ages of ratas were difficult to assess accurately. Increment borings were taken from a number of trees but it was impossible to determine distinct annual rings (see Section 1.3). Age was recorded as the diameter, at breast height, of the 'tree' (rata plus host) and then subtracting the portion of the host tree that was still visible. (For growth form of rata see Section 1.3). Tree health was ranked on a scale of three according to the leaf density of the canopy. Trees rating 3 were in good condition with very few 'light-spaces' in the canopy: trees rated 1 had very sparse foliage and often had dead branches.

Altitude and aspect were measured with an altimeter and compass respectively while soil moisture was measured in the laboratory from samples taken with an improvised soil corer. The corer was a chrome-plated steel pipe (diam. 2.3 cm) with a sharpened edge. The pipe was marked at 10 cm from the edge and the soil core could be removed with a wooden dowel (2 cm diam.). Each tree was sampled systematically at 10 evenly spaced points around the tree approximately 2 m from the trunk. The 10 samples were placed in a plastic bag and weighed, then dried at 60°C for 48 hours(Evans, 1972), and reweighed in the labratory. Soil moisture was expressed as the weight lost as a percentage of the initial weight.

For horizontal dispersion about individual trees, three trees (1,2,18) were chosen by lot from trees with galls. The sampling procedure is outlined in Section II.2. From each sample a subsample of soil (approximately 10 g) was taken for moisture and ash analysis. Soil moisture was determined as before and soil organic matter was recorded as the percentage weight lost after three hours at 450°C (Evans, 1972). Galls in samples were counted and saved for Section II.10

Vertical dispersion was assessed by selectively sampling about another three trees (1,6,8). Five samples per tree were taken with each sample consisting of a single soil core from the 10 x 10 cm soil corer These cores were carefully extruded into 15 cm diameter plastic bags and taken back to the laboratory. In the laboratory the cores were frozen overnight at -10°C and then cut transversely with a large knife into 2 cm-thick discs. The discs were thawed before a small soil subsample (ca. 1 g) was taken for moisture and ash analysis, and roots and galls were assessed (described above).

An analysis of variance was calculated to see if there was a significant difference between the three trees sampled for horizontal dispersion.

Multiple linear regressions were calculated on a Burroughs B6700 computer using the relevant statistical programme supplied.

Multiple linear regressions were calculated to assess the effect of

(1) tree age, health, altitude, site moisture and aspect on the infestation by P.radicis;

(2) root density, soil moisture and organic content on gall density (horizontal dispersion);

(3) root density and depth and soil organic content on gall density (vertical dispersion);

(4) soil moisture and organic content on root density;

(5) soil moisture and organic content on gall density.

The data, however, was not normally distributed and had to be transformed, using Taylor's law (Southwood, 1966).

$$S^2 = ax^{-b}$$

From a log/log plot of the mean and variance from each tree sample 'a' was determined as -12. 'b' was found by substitution in the equation

$$\log S^2 = \log a + b \log \bar{x}$$

and the transformation factor $p = 1-\frac{1}{2}b = -0.66$, indicating a transformation approximately the reciprocal square root of the raw data. Unfortunately this could not be accommodated in the computer programme used and a more common log transformation, usually employed for contagious distributions (Southwood, 1966) was used. Although gaining the required normality, this transformation lost all zero values from the data and limited the total sample size. (Adding a constant to the original data would only have reintroduced the skew.) Root and gall data were both treated in this manner while soil moisture and ash data were considered normally distributed.

Results

P.radicis was found to attack northern rata in all areas investigated Figure 17.

An analysis of variance (Appendix 8) showed that there was no significant difference between the dispersion about the three trees investigated in the study area. The results of the samples were collated and showed that the variance was larger than the means, indicating a degree of clumping in the dispersion of the population (Southwood, 1963). The equation $y = -10.04 + 0.75x_1 + 0.15x_2 + 1.30x_3 + 1.04x_4 = 1.004x_5$ describes the regression plane upon which the independent variables (aspect, site moisture, tree health, tree age, and altitude respectively) lie. TableXIVi gives the correlation coefficients and the partial correlation coefficients for the dependent (infestation) and each independent variable. Tree age and soil moisture show a significant positive correlation to the infestation of P.radicis. Further statistical data is provided in Tables XIVii-v

The equation $\log y = 0.56 + 0.87\log x_1 + 0.04x_2 - 0.90x_3$ describes the regression plane of root density, soil moisture and soil organic matter with respect to gall density. TableXVi shows that gall density (gall no.) is significantly and positively correlated to root density (root weight). Further statistical data is provided in Tables XVii-v.

The equation $\log y = -1.88 + 0.01x_1 + 0.82x_2$ describes the regression of soil moisture and soil organic content on root density. Soil organic matter is the more significantly correlated factor. Further statistical data is provided in Tables XVI i-v.

The equation $\log y = 0.93 + 1.06x_1 + 0.02x_2 + 0.01x_3$ describes the regression of root density, soil depth and soil organic matter on gall density. Root density and soil organic matter are

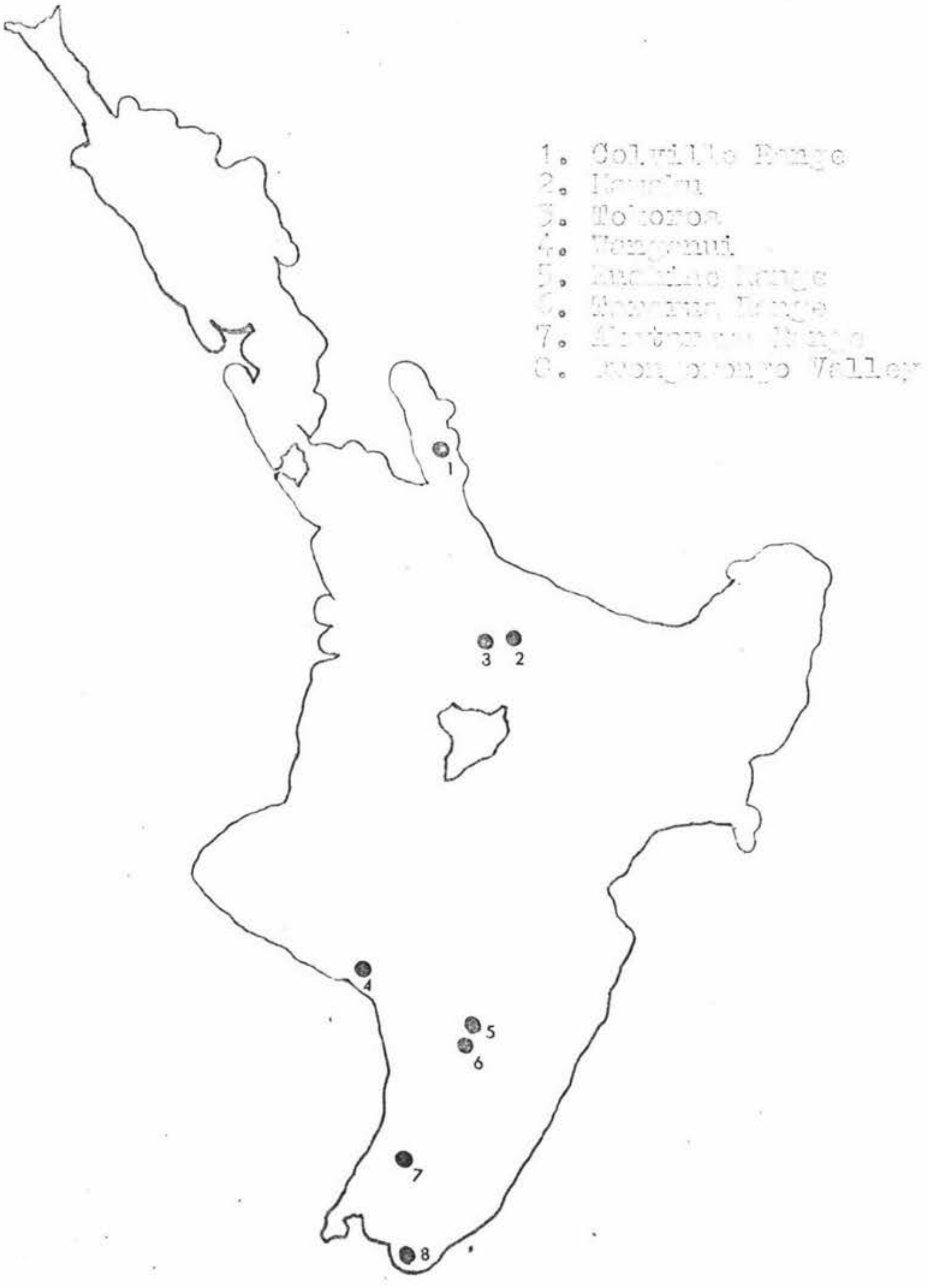


Fig. 17 Map of the North Island showing the areas sampled for P. radicis.

i
```
           CORRELATION MATRIX-
              ASPECT      MOISTURE %     HEALTH       AGE        ALTITUDE    INFESTATION
ASPECT       1.0000       0.6594       -0.4884      0.5718      0.3578      0.5722
MOISTURE %   0.6594       1.0000       -0.3511      0.4818      0.4752      0.6912
HEALTH       0.4884      -0.3511        1.0000     -0.4433     -0.7556     -0.3408
AGE          0.5718       0.4818       -0.4433      1.0000      0.3145      0.7693
ALTITUDE     0.3578       0.4752       -0.7556      0.3145      1.0000      0.4261
INFESTATION  0.5722       0.6912       -0.3408      0.7693      0.4261      1.0000
```

r = 0.623 P = 0.01

ii
```
REGRESSION COEFFICIENTS-

     VARIABLE    COEFFICIENT

     CONSTANT       -10.9354
       ASPECT         0.0747
   MOISTURE %         0.1538
       HEALTH         1.2948
          AGE         1.0381
     ALTITUDE         0.0038
```

iii
```
STANDARD DEVIATIONS AND T VALUES OF
REGRESSION COEFFICIENTS-

      VARIABLE   STD DEVIATION          T     STD DEV BETA        F
        ASPECT          0.6525       0.11           0.2060     0.01
    MOISTURE %          0.0897       1.72           0.2012     2.94
        HEALTH          1.1602       1.12           0.2343     1.25
           AGE          0.2820       3.68           0.1699    13.55
      ALTITUDE          0.0034       1.12           0.2277     1.25
```

iv
```
      VARIABLE     PARTIAL CURR COEF
        ASPECT                0.0306
    MOISTURE %                0.4168
        HEALTH                0.2858
           AGE                0.7013
      ALTITUDE                0.2862
```

v
```
ANALYSIS OF VARIANCE TABLE

    SOURCE      D.F.    SUM SQUARES   MEAN SQUARES        F

REGRESSION        5       120.8296        24.1659      8.52
     ERROR       14        39.7204         2.8372
     TOTAL       19       160.5500

CONFIDENCE LEVEL OF F(    5,    14) =     99.93%
```

Table XIVi-v Correlation data for the infestation of E.robusta by P.pallida.

i

```
             CORRELATION MATRIX-

             ROOT WT.      SOIL MT.      SOIL ASH      GALL No.
ROOT WT.     1.0000        0.3430        0.3655        0.6085

SOIL MT.     0.3430        1.0000        0.8858        0.4058

SOIL ASH     0.3655        0.8858        1.0000        0.3247

GALL No.     0.6085        0.4058        0.3247        1.0000
```

r = 0.45 P = 0.01

ii

```
VARIABLE    PARTIAL CORR COEF

ROOT WT.               0.5658
 SOIL MT               0.2945
SOIL ASH              -0.1921
```

iii

```
REGRESSION COEFFICIENTS-

    VARIABLE    COEFFICIENT
                     -
    CONSTANT      0.5641
    ROOT WT.      0.8664
     SOIL MT      0.0393
    SOIL ASH     -0.9036
```

iv

```
STANDARD DEVIATIONS AND T VALUES OF
REGRESSION COEFFICIENTS-

          VARIABLE    STD DEVIATION         T      STD DEV BETA        F
          ROOT WT.           0.2577      3.36            0.1649    11.30
           SOIL MT           0.0260      1.51            0.3306     2.28
          SOIL ASH           0.9423     -0.96            0.3337     0.92
```

v

```
ANALYSIS OF VARIANCE TABLE

     SOURCE      D.F.    SUM SQUARES   MEAN SQUARES        F

REGRESSION        3         6.1325         2.0442       6.18
     ERROR       24         7.9367         0.3307
     TOTAL       27        14.0692

CONFIDENCE LEVEL OF F(    3,   24) =      99.71%
```

Tables XVi-v Correlation data for the horizontal distribution of galls about individual trees.

i

```
              CORRELATION MATRIX-


             SOIL MT         SOIL ASH        ROOT WT.
SOIL MT.     1.0000          0.7174          0.4060

SOIL ASH     0.7174          1.0000          0.4521

ROOT WT.     0.4060          0.4521          1.0000
```

r = 0.38   P = 0.01

ii

```
HIGH ORDER PARTIAL CORRELATION CO

    VARIABLE    PARTIAL CORR COEF

     SOIL MT                0.1314
    SOIL ASH                0.2526
```

iii

```
REGRESSION COEFFICIENTS-

   VARIABLE    COEFFICIENT

   CONSTANT        -1.8835
    SOTL MT         0.0114
   SOIL ASH         0.8154
```

iv

```
STANDARD DEVIATIONS AND T VALUES OF
REGRESSION COEFFICIENTS-

         VARIABLE   STD DEVIATION          T     STD DEV BETA          F
          SOIL MT          0.0131       0.87           0.1936       0.76
         SOIL ASH          0.4764       1.71           0.1936       2.93
```

v

```
ANALYSIS OF VARIANCE TABLE

     SOURCE     D.F.    SUM SQUARES   MEAN SQUARES         F

REGRESSION        2          3.8929         1.9465      6.00
     ERROR       43         13.9548         0.3245
     TOTAL       45         17.8478

CONFIDENCE LEVEL OF F(     2,    43) =        99.50%
```

Tables XVI i-v Correlation data for the horizontal distribution of roots about individual trees of M. robusta.

i

```
CORRELATION MATRIX-

          ROOT WT      DEPTH      ASH %      GALL NO.
ROOT WT.  1.0000     -0.1692     0.5062     0.9154

DEPTH     0.1692      1.0000    -0.4390    -0.1408

ASH %     0.5062     -0.4390     1.0000     0.6660

GALL NO.  0.9154     -0.1408     0.6660     1.0000
```

r = 0.43 P = 0.01

ii

```
VARIABLE    PARTIAL CORR COEF

ROOT WT.              0.9091
   DEPTH              0.3769
   ASH %              0.6586
```

iii

```
REGRESSION COEFFICIENTS-

   VARIABLE    COEFFICIENT

   CONSTANT        0.9236
   ROOT WT.        1.0605
      DEPTH        0.0159
      ASH %        0.0103
```

iv

```
STANDARD DEVIATIONS AND T VALUES OF
REGRESSION COEFFICIENTS-

      VARIABLE   STD DEVIATION        T     STD DEV BETA        F
      ROOT WT.          0.0859    12.35           0.0622   152.45
         DEPTH          0.0069     2.30           0.0597     5.30
         ASH %          0.0022     4.95           0.0682    24.52
```

v

```
ANALYSIS OF VARIANCE TABLE

    SOURCE     D.F.    SUM SQUARES  MEAN SQUARES         F

REGRESSION       3         17.3101        5.7700    105.79
     ERROR      32          1.7454        0.0545
     TOTAL      35         19.0555

CONFIDENCE LEVEL OF F(    3,   32) =     100.00%
```

Tables XVII i-v Correlation data for the vertical distribution of galls about individual trees.

significantly and positively correlated to gall density. Further statistical data is provided in Tables XVII i-v

Figures 18 and 19 show graphically the relationship between root density and soil moisture and organic content, and gall density and soil moisture and organic content respectively.

Figures 20 and 21 show graphically the relationship between root and gall density, and soil depth.

Figure 22 shows the summation of the root sample results shown as the distribution of the roots of M.robusta about a typical tree. The distribution is relatively even. By treating the root densities as vectors and summing, the resultant vector is of small magnitude and directed 83° west of north.

Treating gall density similarly produces a large resultant vector directed 61° west of north (Figure 22).

Discussion

P.radicis appears to be limited in its distribution only by the range of M.robusta, suggesting a long association of the two species. In the study area the roots of a tree at the highest altitude (Tree 20, altitude 688 m) were infested with P.radicis. The study area, on this evidence, could not be considered to be at the limit of the species' range.

Dispersion within the study area appeared to relate to a number of environmental parameters, although it must be remembered that the sample size was small (20 trees) and that correlation coefficients are not necessarily indicators of causal relationships.

From TableXIVi it can be seen that tree age and soil moisture are closely correlated to infestation ($r = 0.77$ & 0.69 respectively), although soil moisture is barely significant at the 5% level. By referring to TableXIV('correlation matrix'), in which each variable is correlated to the others, the relatively high correlation coefficient of aspect (w.r.t.

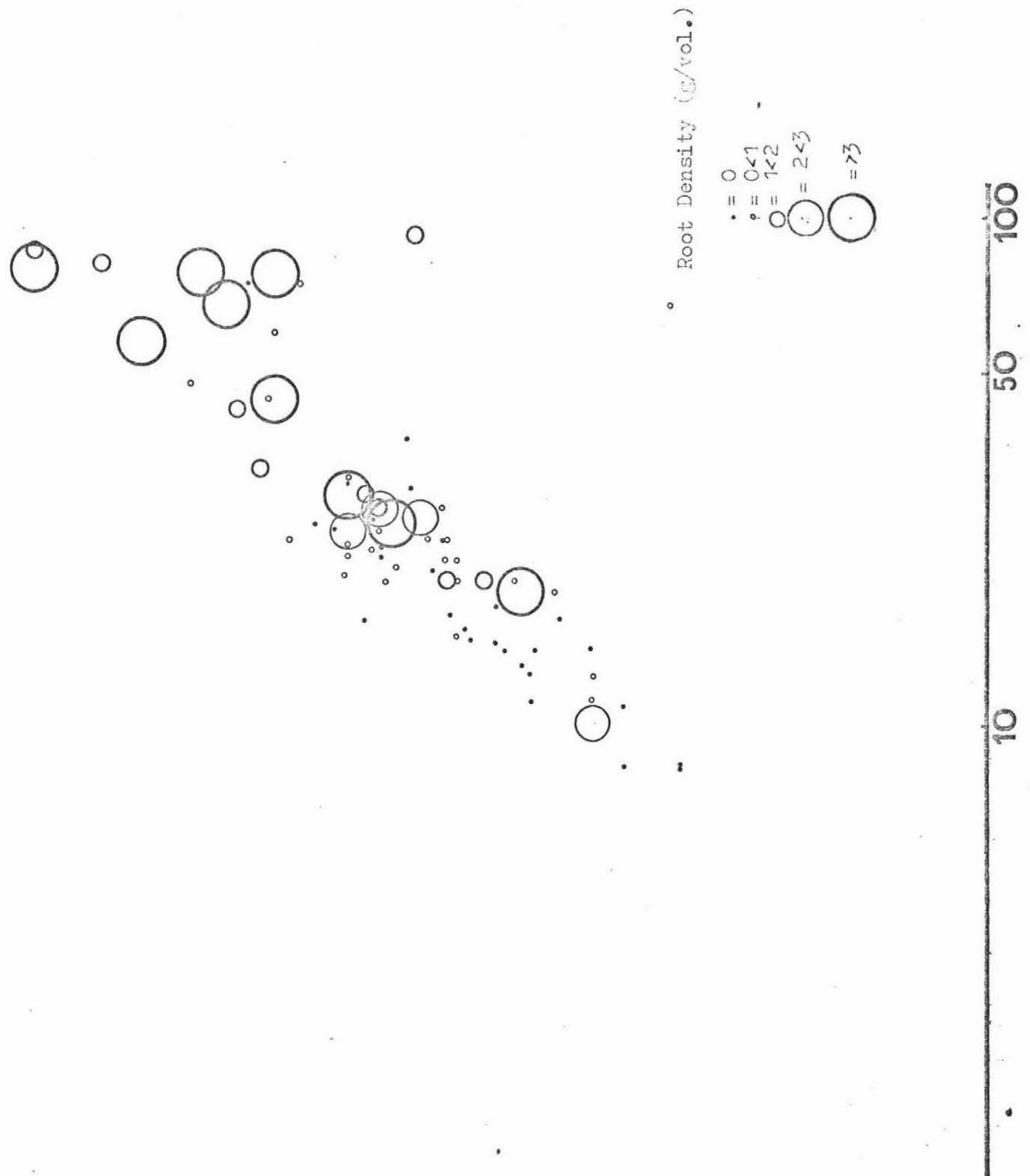


Fig. 18 The relationship between root density, soil moisture and soil organic content.

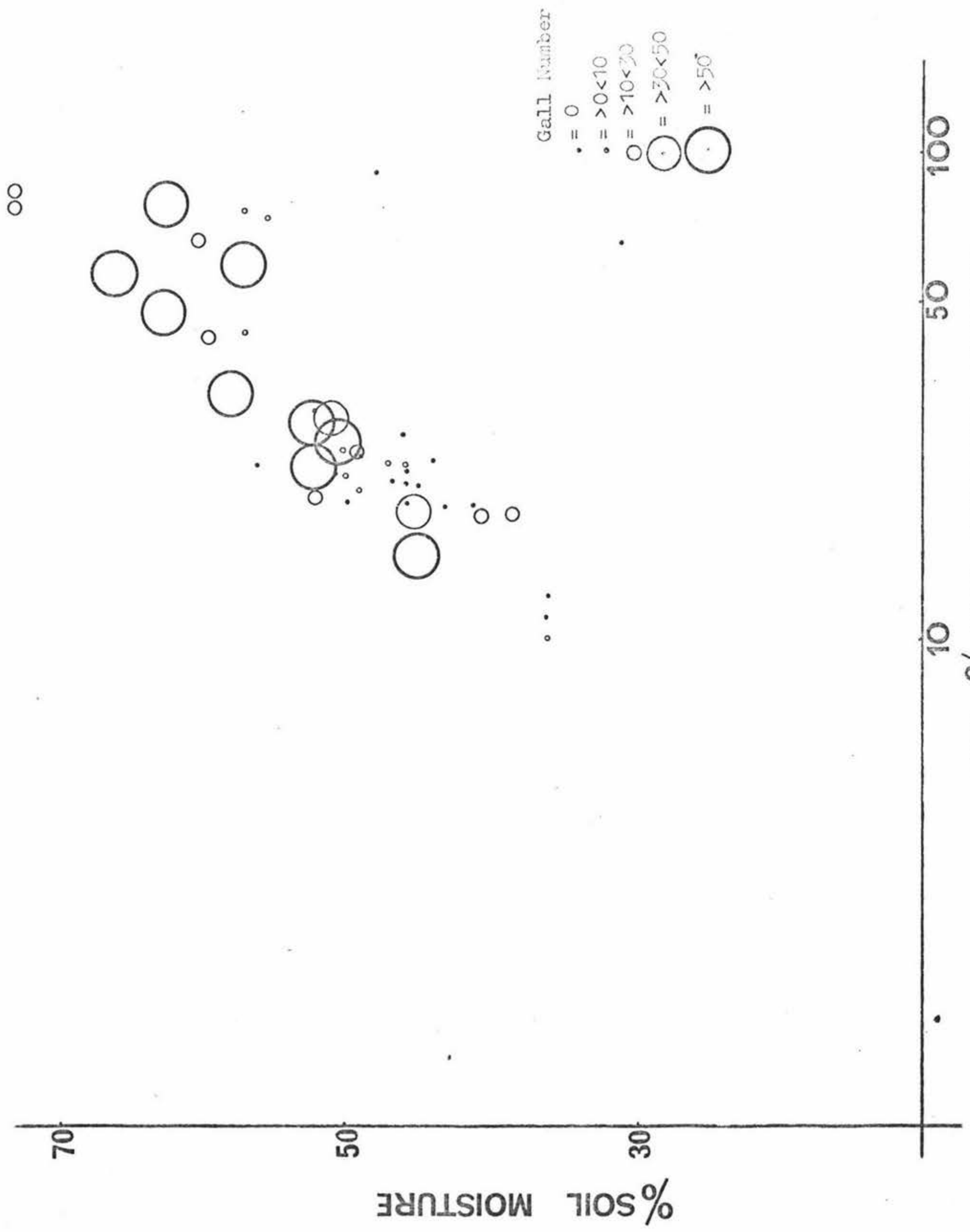


Fig.19 The relationship between gall numbers, soil moisture and soil organic content.

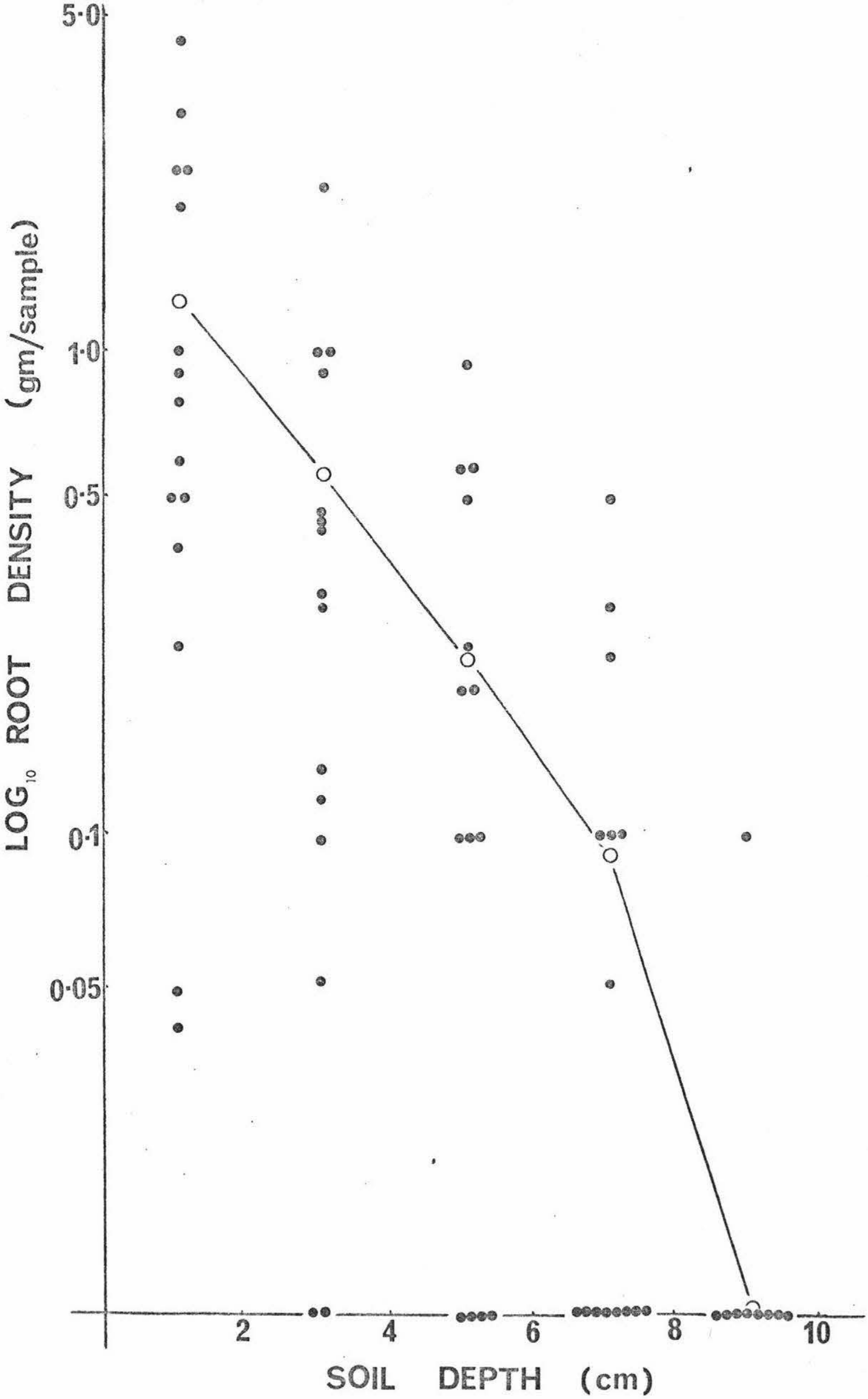


Fig. 20 The relationship between root density and soil depth.

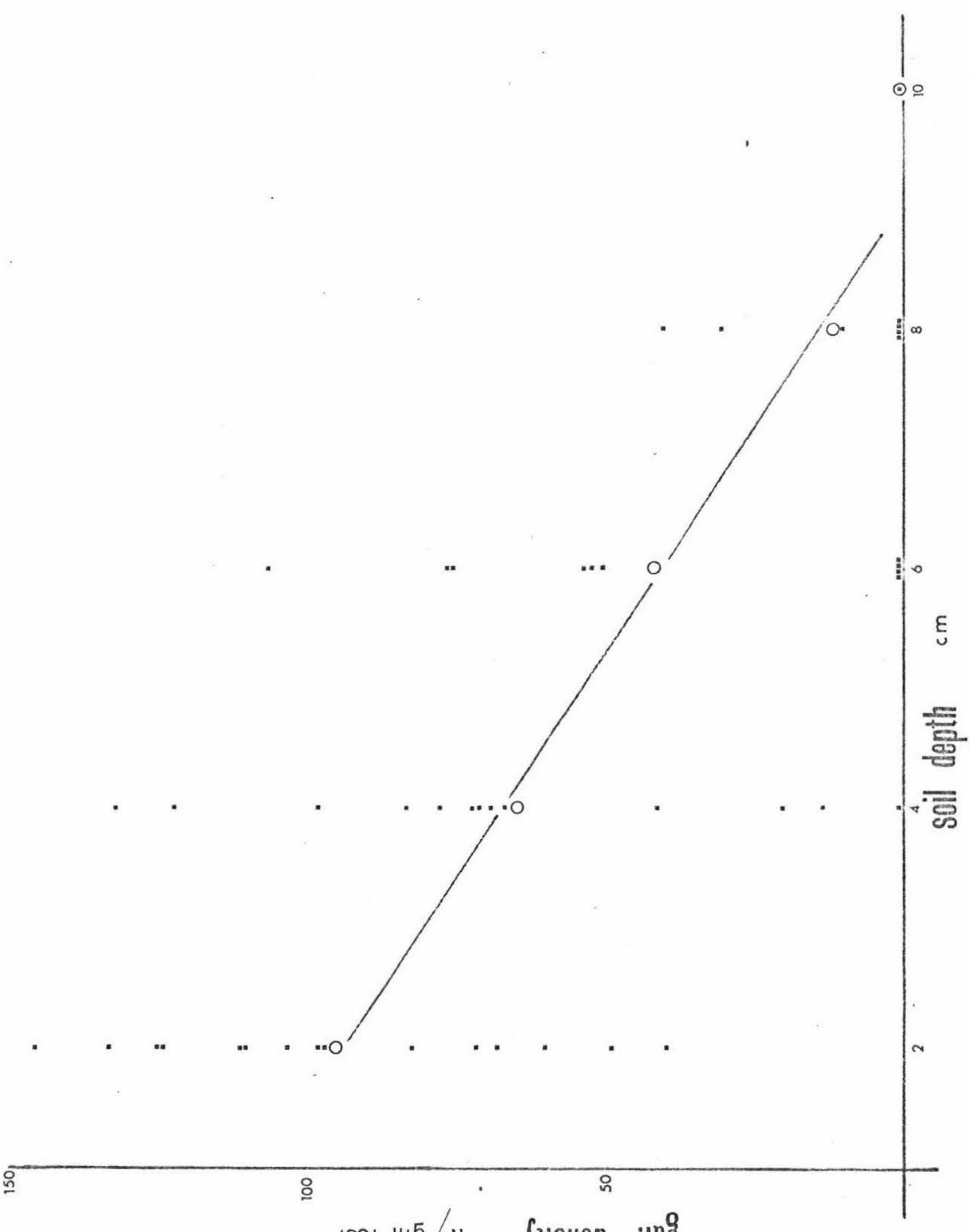


Fig. 21 The relationship between gall density and soil depth

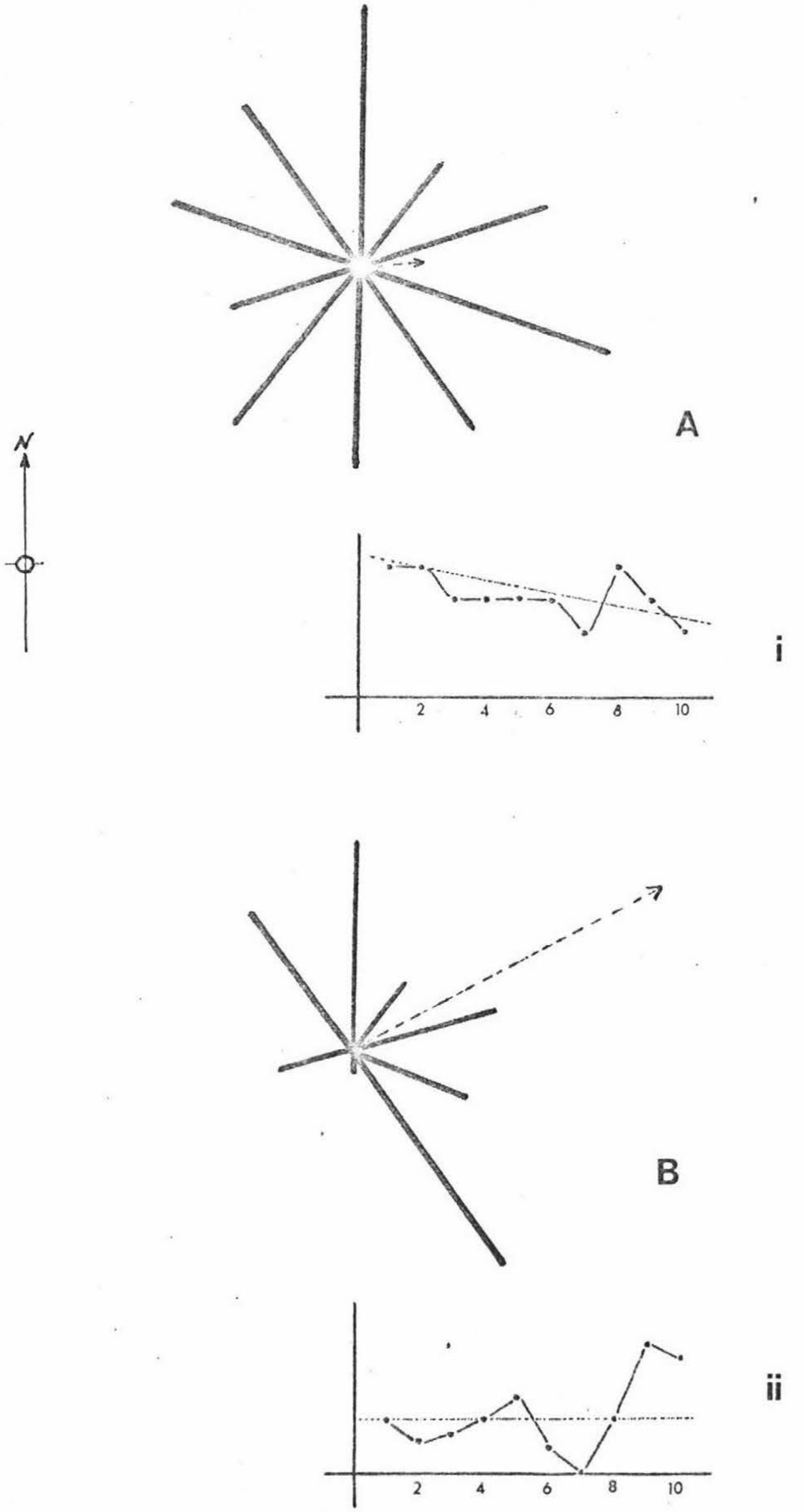


Fig. 22 The distribution of a), roots and b), galls about a typical tree.
i & ii show root and gall density (resp.) with distance from the tree.

-----> = resultant vector

infestation) (r = 0.57) is explained by a close correlation with soil moisture (r = 0.66), confirming the common observation that south-facing slopes in the Southern Hemisphere are damper than north-facing slopes.

It appears then that tree age and soil moisture have some influence on the distribution pattern of P.radicis in the study area. It is a common observation that trees decline in their ability to resist parasites and pathogens as they become over-mature (Kozlowski, 1971): however, the interpretation of age from d.b.h. measurements must be accepted with reservation. Trees of similar age can vary greatly in diameter depending on their site conditions and inter- and intra-specific competion (Daubenmire, 1968). Druce (1966) dated two fallen M.robusta of 3.05 m and 2.45 m d.b.h. at Mt Egmont as being 291 and 331 years old respectively. The life span of M.robusta is not known but Wardle (1971) considers the life span of M.umbellata to be 400-500 years. On these criteria, even accepting a reasonably large error, most of the trees in the investigation were mature or over-mature trees.

A factor which may have biased the results to some extent was the sampling method. As stated above, an accurate measure of infestation would be tedious and time consuming. The method used allowed a quick analysis of the infestation but because of the distribution of roots about a tree (SectionI.3) it may have favoured older trees with higher infestations. For example, an old tree has a more extensive root system than a young specimen. Sampling would therefore be closer to the trunk where root density is higher and there is a greater chance of encountering galls. The extent of the bias is uncertain; most trees were sampled within a 3 m radius from the trunk. Although increased debility is often correlated to increasing tree age, P.radicis infestation was definitely not restricted to old trees and the results of the analysis were not expected from general field observations.

The correlation of dispersion with soil moisture reinforces the observations of Section on the desiccation rates of larvae and roots, and is discussed further below. Other interesting results from Table could be taken into account in any future study of M.robusta. Tree health, for example (ranked 1-3 for poor-good), shows a negative correlation with all parameters measured, i.e. tree health decreases with increased age, soil moisture, altitude and P.radicis infestation. From this one would expect to find healthy trees in dry lowland areas and this was observed in areas adjacent to the study area (Section I.3) and during travels in the North Island. The negative correlation between health and altitude is especially marked ($r = 0.76$) and resembles observations of other species in the Ruahines (Elder, 1965; Franklin, 1967; Ogden, 1971).

The Tables XVi+ii (correlation matrix) show that gall density is significantly and positively correlated to root weight (i.e. root density) ($r = 0.610$) and to a lesser extent to soil moisture ($r = 0.410$). The partial correlation corfficient shows that root density is the more significant factor, and that the soil moisture reading in Table XVi is probably due to a correlation between moisture and root weight, and to some extent, soil organic matter. There is a high correlation between soil organic matter and soil moisture ($r = 0.886$) and this is illustrated in Figures 18 & 19.

The Tables XVI i-v show the results of a multiple regression of soil moisture and soil organic content on root density. Both factors showed a significant positive correlation, with soil ash being more marked than soil moisture ($r = 0.45$ & 0.41 respectively). A similar plot of gall number against soil moisture and organic content does not show the same degree of correlation because of the greater association of galls to roots.

From the regression of soil depth, root density and root organic matter (Tables XVII i-v) it can be seen that again there is a significant positive correlation between gall number and root density (r = 0.915). A slightly less significant correlation (4 = 0.666) exists between gall number and soil organic content. The latter correlation is explained largely by the correlation between roots and soil organic content (partial correlation coefficient). The correlation matrix shows a negative, but not significant, correlation between gall number and soil depth. This is more distinct if gall density (galls/g root) is plotted graphically (Figure 21). A similar graph of root density and soil depth is seen in Figure 20. The loss of zero readings during the log transformation of the data probably produced the less-marked statistical correlations. The negative correlation (r = 0.437) between soil depth and ash, which is common to most soils, probably accounts for some of the fall-off in root density with soil depth.

In review, bearing in mind that the analysis was done with the measurements of root weights and possibly subject to some error, it appears that the small 'feeding' rootlets of M.robusta are confined to the upper 'nutritive' layers of the soil. The organic matter in these upper layers is probably responsible for supplying nutrients and retaining soil moisture. This habitat is generally suited to the needs of P.radicis although it has a tendency to prefer moister regions within this environment. The invasion by P.radicis is limited to some extent by the depth of rata roots. This may be a physical penetration problem or it may reflect difficulties in gas exchange within a deep-soil environment. Sampling in this study was limited by the methods employed in the identification of the host's roots. It now appears that if some error is accepted, a more expansive sampling programme, with greater statistical credibility, could be instigated.

The results from the vector summation of root and gall density about a tree show that galls have a tendency to concentrate in the north-west region of a tree's root-system. This could be explained by the dispersal of adult females prior to oviposition. (Johnson (1969) states that cecids tend to gather on the leeward side of obstacles when airborne). The winds in the study area are predominantly westerlies (Elder, 1958) and the observed distribution could well result from these winds and the dispersal behaviour of P.radicis adults. Figure 22 shows that there is a gradual decline in root density with distance from the tree. A similar plot (Figure 22) for gall density, however, shows that infestation of the roots is relatively constant.

Dispersal

Dispersal is considered here in terms of adult movement to or from the population. Secondary dispersal, i.e. the movement of newly hatched larvae (Hanson, 1959), is considered minimal for P.radicis because of the size of the first larval instar in relation to the host. Adult cecids, although small, are readily dispersed by air currents. The fragile midges have broad, hairy wings and in still conditions they are capable of relatively strong flight (*Mayetiola destructor* recorded 70 cm/sec, Johnson, 1969). In moderate air currents they tend to congregate on the leeward side of obstacles and many have been captured while resting on sheltered spider webs (Johnson, 1969). Gall midges have been recorded at heights of up to 1000 m above ground, but are most numerous below 200 m (Berland, 1935).

Few studies of gall-midge dispersal have been attempted. Walter (1941) recorded *Contarina sorghicola* as flying with prevailing winds across sorghum fields in Texas. Wiseman *et al*. (1972) showed that capture of *C.sorghicola* decreased significantly with distance from the host crop. Harding (1965) found that crop infestation for the same midge was usually from short-, rather than long-, distance migration. Heterogenous dispersal has been recorded in *Mayetiola destructor* where the female travels further than the male (Johnson, 1969). The adult dispersal of *P.radicis* was studied to determine its effect on the population's dynamics.

Materials and Methods

All observations were made in the study area. Malaise trap catches (approximately fortnightly) throughout the year were supplemented with daylight catches from an aspirator and a sweep net (45 cm diam.) during the flight season of *P.radicis*. A small electric suction trap was also in use during the flight season in an effort to determine nocturnal flight

Dispersal

Dispersal is considered here in terms of adult movement to or from the population. Secondary dispersal, i.e. the movement of newly hatched larvae (Hanson, 1959), is considered minimal for P.radicis because of the size of the first larval instar in relation to the host. Adult cecids, although small, are readily dispersed by air currents. The fragile midges have broad, hairy wings and in still conditions they are capable of relatively strong flight (Mayetiola destructor recorded 70 cm/sec, Johnson, 1969). In moderate air currents they tend to congregate on the leeward side of obstacles and many have been captured while resting on sheltered spider webs (Johnson, 1969). Gall midges have been recorded at heights of up to 1000 m above ground, but are most numerous below 200 m (Berland, 1935).

Few studies of gall-midge dispersal have been attempted. Walter (1941) recorded Contarina sorghicola as flying with prevailing winds across sorghum fields in Texas. Wiseman et al. (1972) showed that capture of C.sorghicola decreased significantly with distance from the host crop. Harding (1965) found that crop infestation for the same midge was usually from short-, rather than long-, distance migration. Heterogenous dispersal has been recorded in Mayetiola destructor where the female travels further than the male (Johnson, 1969). The adult dispersal of P.radicis was studied to determine its effect on the population's dynamics.

Materials and Methods

All observations were made in the study area. Malaise trap catches (approximately fortnightly) throughout the year were supplemented with daylight catches from an aspirator and a sweep net (45 cm diam.) during the flight season of P.radicis. A small electric suction trap was also in use during the flight season in an effort to determine nocturnal flight periodicity. The 20 cm diameter trap incorporated a 19 cm plastic, two-bladed, propellor driven by a 6 volt 'Claxon' motor. This was coupled to a 6 volt D.C. car generator belt-driven by a McCollough 2-stroke motor. Beneath the fan a bolt-silk cone directed the catch into a jar of alcohol which could be replaced without stopping the fan. The generator was placed about 30 m from the trap and its output could be read from a voltmeter and adjusted by altering the idling speed.

To investigate immigration and emigration from a population about a particular tree a number of 'fly-paper' traps were used. Each trap was made from a stake about 1.5 m long supporting two 15 cm cross-arms 1 m apart. The stake was hammered into the ground until the lower cross-arm was at ground level. Two plastic sheets (0.15 x 1 m) were greased with chassis grease on one side and clipped to the cross-arms - one to each side - with greased surface outermost.

Two trees (3,6) were chosen for convenience and 8 traps were placed about each. The roots of rata were known to extend some distance from the tree (SectionI.3). Four traps were placed 10 m from each tree, one at each cardinal point. The other four were placed 20 m from each tree at inter-cardinal points. It was considered that the catch from the 10 m traps would indicate emigration while that from the 20 m traps would indicate immigration of P.radicis. Flying insects alighting upon, or colliding with, the greasy surface were trapped. The traps were left in the field for two weeks. The sheets were then folded on themselves and brought back to the laboratory where they were washed in petrol which removed the grease so that the catch could be examined.

Results

None of the traps used recorded any flight activity for P.radicis (Data Appendix 9). Aspirator and sweep-net catches consisted of male P.radicis only.

Discussion

Unfortunately most methods produced negative results, i.e. either no P.radicis adults were captured or the results could not be analysed.

The Malaise traps, although catching numbers of cecids, failed to catch P.radicis. Similarly the suction trap, although catching cecids, did not record any flight activity for P.radicis. Negative results from these methods are difficult to analyse accurately. The traps may have been sited wrongly, although this is difficult to accept for one Malaise trap, as it was sited under a rata tree which carried a large number of galls (Tree 8, infection rank 7/10). The suction trap, placed under Tree 3 in 1972 was shifted to Tree 1 in 1973. The 1973 run, however, was plagued by mechanical failure and only a few short runs were possible.

The fly-paper traps unfortunately suffered from rain and wind damage and after laboratory methods to clean the specimens, small insects were too fragmentary to be identified. Aspirator and sweet-net catches (daylight), however, revealed an interesting trend. All catches (some 50 adults) were of male P.radicis only. Females caught in the field had to be 'dug' from the litter - as in laboratory rearing cages (Section II.4).

The fact that other cecids were caught indicates that the traps were potentially reliable. If sitings were adequate it can only be assumed that P.radicis has poor dispersal capabilities. Males were found resting upon twigs, spider webs, fern etc. close to the ground. Sweeps with the net were only successful if these structures were disturbed in the process. This habit of clinging to sheltered objects (recorded for other cecids by Felt, 1925) and the cryptic nature of the female may well account for the negative results recorded. The cryptic nature of the female, however, cannot be absolute as a number of females were caught on the sex-attraction trap used in Section II.4. A t-test between males and females caught on control traps revealed no significant difference between the two.

It appears, from the results available, that the dispersal of P.radicis may not be a significant factor in the population's dynamics. Any dispersal that is effected is probably strongly influenced by the prevailing winds of the area. Because of the cryptic nature of the females, dispersal may be more marked for the males of the species. Such a heterogenous dispersal, coupled with the male's polygamy, would tend to increase the gene pool, rather than affect the dynamics, of the total population. However, in an area of low host-plant density (now typical of many of the remaining indigenous forests), most emigrants would be lost to the population. The lack of a strong dispersive stage in the life cycle of P.radicis could be an important factor in speeding any decline in the total population. Sub-populations about individual trees would be lost when the tree died, while the colonisation of young host trees, necessary for the procreation of the species, would be extremely fortuitous.

The Influence of Weather on the Population Dynamics of P.radicis

The weather has an obvious and often direct effect on insect numbers (Andrewartha & Birch, 1954). Cecids, however, once enclosed by plant tissues, are buffered against sudden fluctuations in temperature and humidity. When isolated from the gall the frail insects are susceptible to the vacillation of the weather. The adults, eggs and first larval instar of P.radicis do not live within the gall. The three stages are concurrent and overlapping and adverse weather could affect all three simultaneously. The stages are compressed into a period during which weather conditions are normally very favourable (Figure 3), i.e. the weather is calm with temperatures high and rainfall generally adequate to prevent serious desiccation of the forest floor. Recently, however, the Manawatu District has experienced two droughts (Section I.2). The drought of 1972-73 continued almost to the end of flight period of P.radicis and many partially exposed roots of M.robusta were desiccated.

Adults of P.radicis were probably not affected by thse conditions to the same extent as the eggs and larvae. Adults are adapted to aerial conditions and may migrate short distances to a more favourable micro-environment. Their life span is so short that their biological function is probably concluded in 1-2 days anyway. (The adults appear to be susceptible to cool temperatures (Section II.4). In contrast, eggs and larvae are probably more susceptible to drought conditions. Eggs are undoubtedly protected from desiccation to some extent by the chorion (Imms, 1957). The small, thinly cuticled larvae, however, have a large surface-area:volume as well as spiracular and alimentary openings and must be prone to water loss. This section attempts to evaluate the effects of drought conditions on first instar larvae, galls and normal roots, in terms of desiccation rates.

Materials and Methods

All plant and insect material was obtained from the study area. It was stored in plastic bags at 4 ±1°C for no longer than 24 hours prior to experimentation.

The 'desiccators' were 4 and 5 decimal place Mettler balances (4 place for galls and roots and 5 place for larvae). The weighing pans of these balances can be isolated from ambient conditions by sliding plexiglass doors. The weighing pan was surrounded by three small open dishes (2.5 cm diam.) containing P_2O_5. A small dial hygrometer was placed alongside the pan and the doors closed. When the relative humidity levelled at about 38% RH (22 ±2°C) one door was opened and the sample quickly placed on the pan. The door was reclosed and the initial weight of the sample was noted. Thereafter the weight of the sample was recorded at intervals of 5 minutes for a total period of 30 minutes. In some instances weighing continued for 16 hours (overnight) at longer intervals.

The balance pans were allowed to hang freely between weighings to minimise errors caused by resetting the balance (resetting could lend to considerable error, especially in the 5 decimal place balance.)

The root material used was, as near as possible, fresh and undamaged. White galls (i.e. galls containing second or early third instar larvae) were cut from roots immediately before the experiment. Eighteen first instar larvae were obtained from three Baermann funnels (Section II.9). Unfortunately all the larvae had to be used at one time as the weight loss for one individual was too small to be measured accurately by the apparatus. All material was washed and 'blotted' dry upon filter paper before weighing commenced. After weighing, the material was dried at 60°C for 24 hours (Evans, 1972) and reweighed. Desiccation was expressed as a percentage of total water content lost with time. The root and gall desiccations were repeated.

Results

The results are expressed in Figure 23 and Appendix 10. The first instar larvae rapidly dehydrate under these conditions (RH 40 ±5% 22 ±2°C) losing 40% of their water content in 15 minutes. No movement was seen after this time and the larvae were presumed to be dead. Normal roots also dehydrate fairly rapidly although a critical dehydration level was not established. Galls containing second instar larvae exhibited the slowest dehydration rate of the three tissues tested.

Discussion

The susceptibility of first instar larvae to desiccation has been demonstrated. However, the significance of dehydration in natural conditions is difficult to assess in terms of larval mortality.
The drought of 1972-73 possibly broke before the larvae had emerged.
The larvae themselves are capable of some locomotion and may be able to move with sufficient rapidity to escape desiccation. It is also possible that the oviposition behaviour of the female is such that eggs, and hence larvae, are not placed in areas susceptible to desiccation (see Dispersion).

The fine rootlets of rata which are attacked by P.radicis have a high surface area:volume ratio and hence susceptible to desiccation. The rate of water loss experienced in the laboratory could probably lead to a critical water loss in a drought period in the field. Galls, on the other hand, are virtually spherical with a low surface-area:volume ratio and a correspondingly low water loss. The very slow dehydration rate observed illustrates the buffering effect the gall provides for the enclosed larva.

In normal conditions it is unlikely that the weather has a marked direct effect on population numbers. The stages of the life cycle of P.radicis which are susceptible to weather are synchronised with normally optimal climatic conditions. The frail adults are probably the most susceptible to temperature fluctuations. The adult life-span, however, is

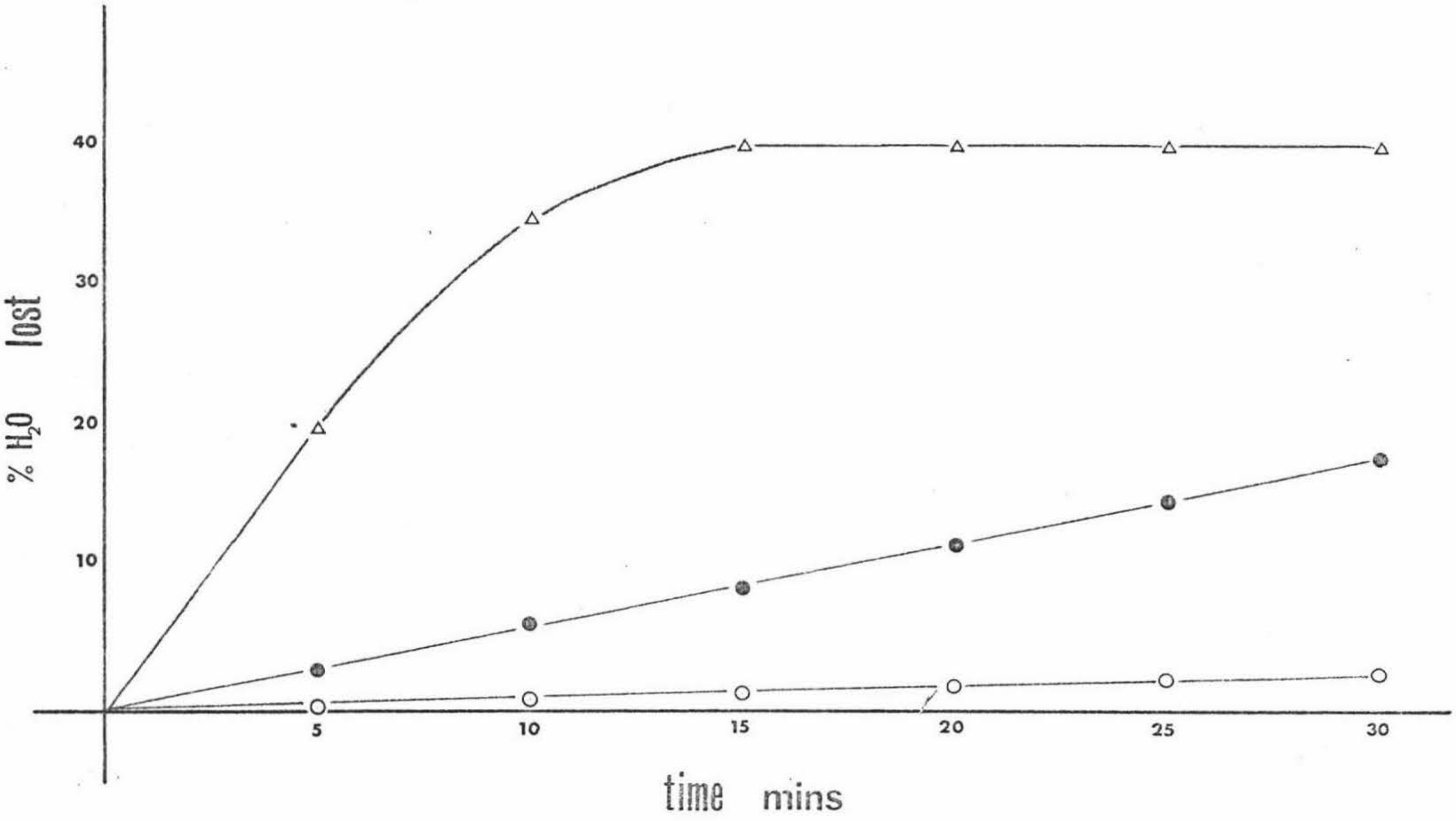


Fig.23 Mean desiccation rates of first instar larvae (△), roots (●) and galls○at 22 C and 40% R.H.

very short and their biological function (i.e. mating and laying eggs) is probably concluded during the day of emergence. The flight period of *P.radicis* is approximately 4-6 weeks so that prolonged adverse weather conditions would be necessary to produce a marked effect upon the population. Indirectly the weather may limit the feeding sites of *P.radicis* larvae by significantly reducing the root biomass of *M.robusta*. This loss, coupled with the degree of competition for gall sites (Section II8) could well be an important factor in limiting the population.

Intra-specific Competition

No concerted effort was made to assess competition. Nearest-neighbour techniques (Southwood, 1966), often applicable to galls or stationery insects of aerial plant roots, were too difficult to apply to the finely-divaricating, three-dimensional root system. Any measurement or classification of the root system is hindered by the root's lack of definable spatial structure and virtually continuous growth.

Materials and Methods

The methods used were essentially an extension of the root growth study of Section II.3 (Rata Phenology). The same five samples per tree were used and counts of root tips were supplemented with counts of galls. The study ran from 4.10.72 to 3.3.73. Some selective samples were taken during, and outside, this study period.

Results

Counts of root tips and galls from samples taken during the study period are collated in Table XVIII.

Discussion

From Table XVIII it can be seen that in no instance did the root-tip-to-gall ratio equal one, let alone approach zero, i.e., in all samples the root tip count exceeded the number of galls. Even in selective samples of high gall densities (Plate 18) a number of ungalled roots were present. This method, however, essentially measures the presence of successful larvae - possibly the results of competition. A comparison of the estimated number of eggs laid/g of root (90/g, Section II.5) by adults, and the observed root-tip counts (tips + galls) shows that the highest average number of root-tips (39.16/g) is less than half the required figure. This suggests a very strong competition for a gall site. A restriction in the number of feeding sites could well prolong the duration of the first larval

Sample date	Av. wt (g)	Av. no. root tips	Av. no. galls	tip:gall
4.10.72	4.42	137	20	6.85
10.11.72	6.32	153	19	8.05
21.12.72	3.87	102	27	3.78
14.1.73	4.60	98	17	5.77
5.2.73	5.83	146	11	13.27
3.3.73	5.95	201	32	6.28

TABLE XVIII THE RESULTS OF COUNTS OF ROOT TIPS AND GALLS FROM SAMPLES TAKEN FROM THE STUDY AREA

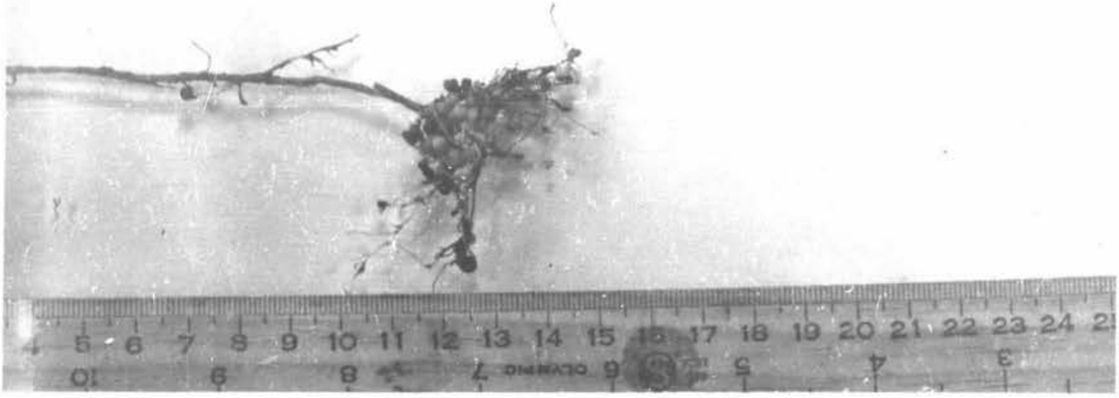


Plate 13 An example of a high gall density.

instar. In the laboratory eggs were laid in clumps or singly, on or off, root material. The degree of selection of a gall site by the female is not known but the first instar larvae are equipped for locomotion (SectionI.4), and have been observed moving freely along roots. Competition, reflecting the time taken in finding a gall site, must determine to some extent the susceptibility of first instar larvae to predation.

At the feeding site the larvae induce a substantial growth of plant tissue which is necessary for the normal development of the insect. The ability of the plant to supply metabolites for gall production during heavy simultaneous invasion by first instar larvae was not investigated. Although dense infestations were encountered it was not known whether these resulted from simultaneous or staggered attack. If the availability of metabolites was limited, the first larval instar could be prolonged with a subsequent increase in the chance of predation. Competition for nutrients between larvae in established galls was not determined although respiration studies of galls (SectionIII.4) indicated that gall metabolism is generally lower than normal root metabolism, and the nutrient demand of larvae may be so low that competition does not arise. There was no obvious limiting 'individual-distance' between galls and intense clustering was not uncommon.

The presence of high gall densities (Plate 13) suggests that lateral roots may be initiated in response to the larvae feeding or, more likely, to compensate for the galling of adjacent roots. The premature death of young rootlets is a common occurrence in many trees (Kozlowski, 1966), and presumably these trees are adapted to compensate for this loss. Torrey (1950) has shown that root-tip decapitation results in a marked increase in the initiation of lateral roots. Although the root meristem is not lost until the third larval instar, the 'functional decapitation' inflicted by the larvae of P.radicis may lead to a greater initiation of lateral roots in an infested region. Torrey (1959) showed that the initiation of lateral roots was controlled by a complex of factors, not all of which were synthesised in the root. A limited supply of these substances (IAA, nicotinic acid, thiamine, etc.) may be responsible in suppressing root initiation.

Mortality - predation

To some extent the juvenile stages of the gall midge may be protected from their enemies by the structure of the gall. However, a wide range of predators and parasites have been recorded as attacking gall midges. The range includes other Cecidomyiidae, Heteroptera, Coleoptera, Hymenoptera, Acarina, Nematoda, fungi and a number of vertebrates (Mani, 1964; Felt, 1925). In some instances the gall midge may be an incidental victim of a predator feeding on the nutritious gall.

The mortality of gall insects can often be inferred from the absence of the insect or the presence of a predator or parasite. In some cases the type of damage done to the gall can give a clue to the identity of the predator. The predation or parasitism of those stages of the gall insect which are isolated from the gall is more difficult to establish.

For the greater part of its life cycle *P.radicis* inhabits a soil environment abounding in eurytopic predators. It was considered unlikely that a virtually immobile, soft-bodied insect would escape some degree of predation or parasitism in such an environment. In this section the mortality of the *P.radicis* population is assessed and some effort is made to identify the causal agents. Predation and parasitism are considered separately and nematodes, because of the extraction techniques, are considered as predators.

Predation

Materials and Methods

During the normal fortnightly samples (Section II, Life cycle) mortality was measured from the presence of galls containing the remnants of larvae or pupae, and galls in which normal root growth had resumed. The latter were characterised by the root recovering from the spiral growth caused by the presence of the larvae in the initial stages of gall formation. In the later stages or after completion of the gall the death of the larva resulted in a straight normal root growing through the side of the gall. All galls showing this character were white and typified the galls of the second larval instar. (Empty brown galls with exit holes cut were considered to be galls of a previous generation.) The remains of third instar larvae were usually present within a gall after death. The remains of second instar larvae, however, were never found.

Possible predators of *P.radicis* were identified by a species association technique (Southwood, 1966). The experiment was primarily designed to elucidate predators of the egg and first larval instar and sampling was done two weeks after the termination of the flight period (21.3.73). However, as second and third larval instars are also present at this time the correlation can be extended to include them.

Five random samples were taken at each of five randomly chosen trees on the north bank of the study area. Samples were randomised by the method outlined in Section : however the area was limited to within 5 m of the trunk (i.e. concentric points were 0.5 m apart, see p. 28-29). sample consisted of two adjacent cores from the 10 x 10 cm soil cover. The cores were labelled, packed in plastic bags and brought back to the laboratory. One core from each sample was searched for arthropods and the other for nematodes.

Arthropods were extracted from samples by means of Berlese funnels (25 x 25 x 40 cm, 40 watt bulk) over a period of three days: 13 funnels were in use and those samples not accommodated in the first extraction were stores in plastic bags under dark conditions at $4 \pm 2^{\circ}$C. The whole core sample was used for the extraction. Arthropods migrated downwards to escape desiccation and were collected in a beaker of alcohol at the base of the funnel.

Nematodes were extracted by means of seven Baermann funnels. Those cores not used immediately were stored in plastic bags at 4 ±2°C. Only a 50 g subsample was used for nematode extraction. This was placed in a 250 ml beaker which was closed with a square of cotton gauze. The beaker was filled with water and upturned in a 10 cm diam. plastic funnel. The water level in the funnel was adjusted to approximately 1 cm above the gauze. Extraction continued overnight for about 24 hours. Nematodes migrated downwards and collected in the stem of the funnel whence they could be released into a beaker.

After extraction the soil cores from the Berlese funnels were inspected for the presence or absence of galls. The result of the observation was used for both 'nematode' and 'arthropod' soil cores. First instar larvae of P.radicis were also found to be extracted from Baermann funnels. Their presence in cores tested for nematodes was also recorded as a presence in 'arthropod' cores.

After extraction, arthropods were stored in alcohol until they could be identified. Nematodes were identified immediately after extraction by means of an improvised key (Appendix II). The nematodes from each sample were collected in a petri dish from which they were removed with a fine, hooked, pin and placed in a drop of water on a microscope slide. The slide was gently heated over a spirit burner until the nematodes were immobilised. The water was drained off and the specimens mounted in glycerine. Specimens were identified as species A, B, C etc. depending on general body size, anterior and posterior characters. No attempt was made to identify the species authentically until after analysis. Arthropods were identified to Class, Order or Family level.

Association analysis was carried out between the presence of galls and larvae (separately) and species or other grouping of animal found in the samples. Fager's (1957) Index of Affinity was used.

$$I\,AB = \frac{2J}{nA + nB}$$

This can be converted to a one-tailed 't-test' where

$$t = \left[\frac{(nA + nB)(2J-1)}{2nAnB} - 1\right]\left[nA + nB - 1\right]$$

and the minimum significant value for t is 1.645 (at the 5% probability level).

nA = the number of occurrences of species A

nB = " " " " " " B

J = " " " joint occurrences

The number of occurrences of A must lie between half and equality to those of B (Southwood, 1966), i.e. a comparison cannot be made between two species if one is less than half as prevalent as the other.

This index is suitable for heterogenous habitats and only measures positive associations.

Results

The percentage of mortality observed in fortnightly samples is recorded in Table XIX

The results of the species association analysis are presented in Table XX. A centipede (Zelanion sp?), trombidiform mites and a nematode (Monochidae?) show a significant association with first instar larvae of P.radicis (Data Appendix 12).

Discussion

The mortality of second and third instar larvae and pupae, as recorded from fortnightly samples, is fairly erratic and no consistent pattern can be seen from month to month. If mortality was constant throughout the year a greater percentage of mortality would be expected per sample with the passage of time. Such an increase was not apparent, although there is a slight rise between the two halves of the year (15 to

Month*	2nd instar larvae	2rd instar larvae	Pupae**	Total
March	9	5.5	1	15.5
April	17.5	3	0.5	21
May	6.5	13.5	1	21
June	1	0	0	1
July	10.5	2.5	1	14
August	12	5	0	17
September	3	9	0	12
October	27	11	22	40
November	11	7	1	19
December	21	10	0	31
January	0	6	0	6
February	17	9	0	26

TABLE XIX THE PERCENTAGE MORTALITY OF SECOND AND THIRD LARVAL INSTARS AND PUPAE OF P.radicis, RECORDED FROM FORTNIGHTLY SAMPLES

* average of fortnightly samples

** % mortality excluding parasitism

INVERTEBRATE	Fager's index (t)	
	Galls	First larvae
ARTHROPODS (excluding ACARINA)		
Crustacean sp. A (AMPHIPODA)	1.5	-
Centipede sp. A (CHILOPODA)	2.88*	1.70*
SYMPHYLA sp. A	1.59	0.23
ACARINA		
Species i (TROMBIDIFORMES)	0.021	0.026
CRYPOSTIGMATA	2.73*	0.74
TROMBIDIFORMES	-	2.04*
PARASITIFORMES	-	-
METASTIGMATA	-	-
NEMATODA		
Sp. A	2.14*	0.91
C	3.65*	2.20*
D	0.34	-
E	0.57	0.44
F	0.34	-

TABLE XX SPECIES ASSOCIATION ANALYSIS -
The species most closely associated with galls
and first instar larvae of P.radicis

* t = 1.645 is significant at the 5% level

24%). This result could be explained if mortality occurred at one time in the year and the samples represented estimates of this. However, this is not likely considering the dynamic state of the population. A more likely explanation would be the disappearance of the evidence of mortality. An empty gall from which a root regenerates, rapidly loses its resemblance to a gall and might not be detected. Similarly a dead larva might decompose and the gall merely be recorded as 'empty'. Empty galls in their turn might be broken down and lost during sampling. It is therefore difficult to make an accurate estimate of mortality in this population where the disappearance of larvae is not adequately determinable by such means as a gain in host weight or increase in volume. As mortality was probably greater than that recorded, the values estimated are probably best regarded as minimal values of mortality within each stage (11.29% and 6.79% for second and third instar larvae respectively).

Mortality in the third larval instar appeared to be almost exclusively associated with a fungal attack (although one mature gall, of the many dissected, did contain a nematode). Whether or not the fungus was the primary, or merely a secondary, agent is not certain. The fungus was devoid of fruiting bodies but the hyphae were septate and it was probably a member of the Ascomycetes.

It was determined in Section II (Dispersion) that roots and galls were generally associated with high soil ash and moisture regimes. These areas are probably the more fertile regions of the forest floor and could be expected to harbour a greater number of organisms. The associations found by this analysis could therefore be incidental, or indirect, in light of other associations in such a fertile environment. It can be seen that many species, in view of their low numbers, could not be used in the analysis (Appendix 12).

Of the arthropods only the centipede (sp. A) had a significant association with either galls or first instar larvae (t = 2.88 and 1.70 respectively; t = 1.645 significant at 4%). All centipedes are predaceous although generally not prey-specific (Borror & Delong, 1964), and this species probably accounts for some mortality, especially of unprotected larvae (i.e. first and early second instar). The crustacean (sp. A - Amphipoda) was present in almost all samples and therefore not significantly associated with galls or larvae. The scavenging habit of the amphipods probably exclude them from effecting any significant mortality of P.radicis. The symphylan (sp. A) could be considered in the same light. Although plentiful it was not significantly associated with galls or larvae. There was a close association with galls and in other sampling programmes symphyla were often found inhabiting old galls.

The hymenopterous sp. A, although failing to be significantly associated with galls or larvae, was in fact the pupal parasite of P.radicis (Section II.10) living free in the soil. Other arthropods, when grouped, e.g. Staphilinidae and Dipterous larvae, although showing a significant association with galls, do not appear to be of significance to larval mortality. However, when mites were grouped, as near as possible, into their respective superfamilies (Kuhnelt, 1961), a significant association was found between first instar larvae and Trombidiformes. Trombidiform mites are generally regarded as predators and are not significantly associated with galls (i.e. plant matter). Cryptostigmata (omnivores), on the other hand, are the reverse; showing a significant association with galls but not the larvae. Mites are typically very numerous in forest litter and are responsible for a large proportion of biomass turnover (Evans, 1955). It is probable that Trombidiforme mites, although not prey-specific, are responsible for some larval mortality.

The nematode analysis is possibly more conclusive. Of the species encountered, only sp. A and C were significantly associated with galls or larvae. Species A showed a significant association with galls ($t = 2.138$) and species C showed a significant association with both galls and larvae ($t = 3.645$ and 2.199 respectively). The mouthparts of the two species correlate closely with the association in which they were found: the pointed stylet of species A typifies a plant sting nematode (Jones, 1965) while the sickled teeth of species C typify a predator. From its general appearance and mouthpart structure, species C is probably a member of the Family Monochidae (Jones, 1965). Immediately following this analysis one nematode (sp. C) was found within the mature gall of a third instar larva. It is probably then that this nematode is of some significance in the mortality of all the larval stages of P.radicis.

The larvae of P.radicis inhabit an environment abounding in invertebrate species. Many of these are predators and some (centipede, mites, and a nematode) are statistically significantly associated with the larvae and must be suspected of causing some mortality, especially of more exposed larvae.

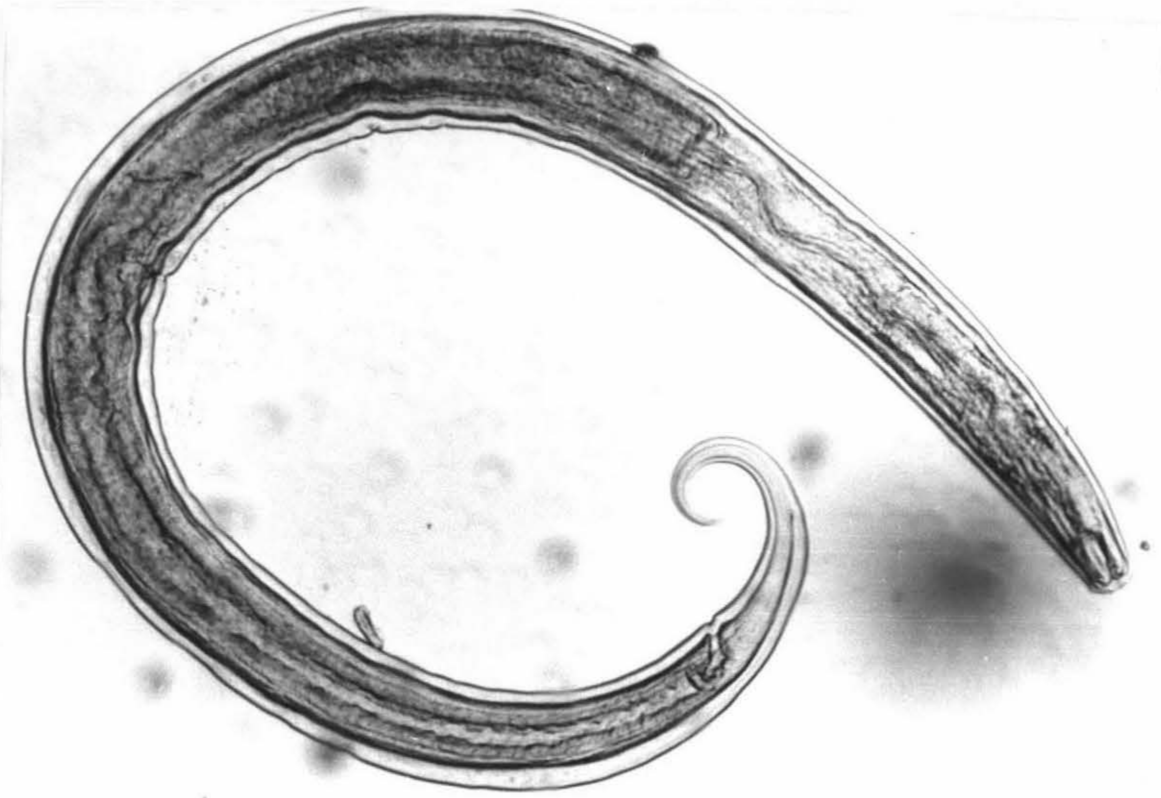


Plate 14 The nematode predator of P. radicis.

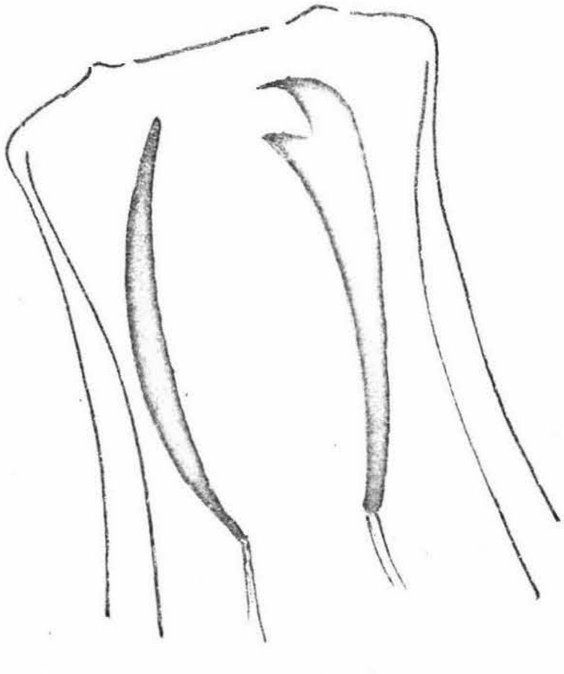


Fig. 26 The mouthparts of the nematode predator of P. radicis.

Mortality - parasitism of P.radicis

Sampling during the summer of 1972 revealed a small hymenopterous parasite of P.radicis. Typically parasites effect age-specific mortalities which may significantly affect the dynamics of the whole population (Clark et al., 1967). The effects of parasites are often cited as examples of density-dependent regulations which act to maintain the stability of the host population (Nicholson, 1933; Nicholson & Bailey, 1935; Utida, 1955, etc.). The parasite in its turn is subject to controls dependent on its life history.

The biology and the degree of mortality effected by the parasite of P.radicis is investigated in this section.

Materials and Methods

The biology and life history of the parasite was recorded as it coincided with normal samplings for the biology of P.radicis.

Parasites at various stages of development were cleared, mounted and measured as described for P.radicis (Section I.4)

A number of adults were reared from pupae dissected from galls. The behaviour of reared adults, when placed in petri dishes with galls containing pupae of P.radicis, was noted.

Three newly emerged adult female parasites were dissected to determine fecundity.

The material for the analysis of the pupal parasitism of P.radicis was obtained from samples investigated in Section II.5. The sampling undertaken during the summer of 1972 showed that the development of the parasite was rapid and restricted to the prepupae and pupae of P.radicis. Sampling in 1973 therefore was undertaken just after the time when the greatest numbers of P.radicis would be in the pupal stage, at which time the greatest amount of parasitism was assumed to have occurred.

Galls, which were stored in alcohol until required, were dissected and their contents recorded. Parasitism was assumed to have occurred if a parasite larva or pupa, or a meconial pellet, was found within the gall. The presence of a cecid pupa, or pupal exuviae was taken as a measure of the pupal population of P.radicis.

Results

The parasite was identified as a member of the Ceraphronidae.

The adults (Figure 24) are small (2mm) brown-winged insects. The marginal vein of the forewing extends from the wing base to about two-thirds the length of the wing. The vein is broken at a point four-fifths of the way along its length. The small distal portion subtends a short arc-shaped vein which forms an unclosed marginal cell.

The ♀ antennae are 10 segmented and geniculate with a long scape. (♂ 11) The tibia of the forelegs bear two combed spurs.

When newly laid the egg is elliptical (0.25 x 0.1 mm) and has a small process at one end (Figure 25).

The larvae are virtually featureless blobs with the only sclerotised structure being a pair of stout, pointed mandibles. The eggs and larvae are semi-translucent. The number of larval instars was not determined.

The eggs of the parasite are laid on the anterior region of the prepupa or pupa of P.radicis as it lies within the gall. At this stage P.radicis lies with its anterior region close to the hole intended for the exit of the adult (Section II.3). In most instances only one egg was found per pupa; however, two per pupa was not uncommon and one pupa was found encrusted with five eggs. The larvae of the parasite were invariably found attached to the abdomen of the pupa of P.radicis.

Towards the end of its larval development the parasite becomes creamy white and opaque. Before pupation one large, shiny-black, meconial pellet is extruded and lies in the hollow of the curved prepupa

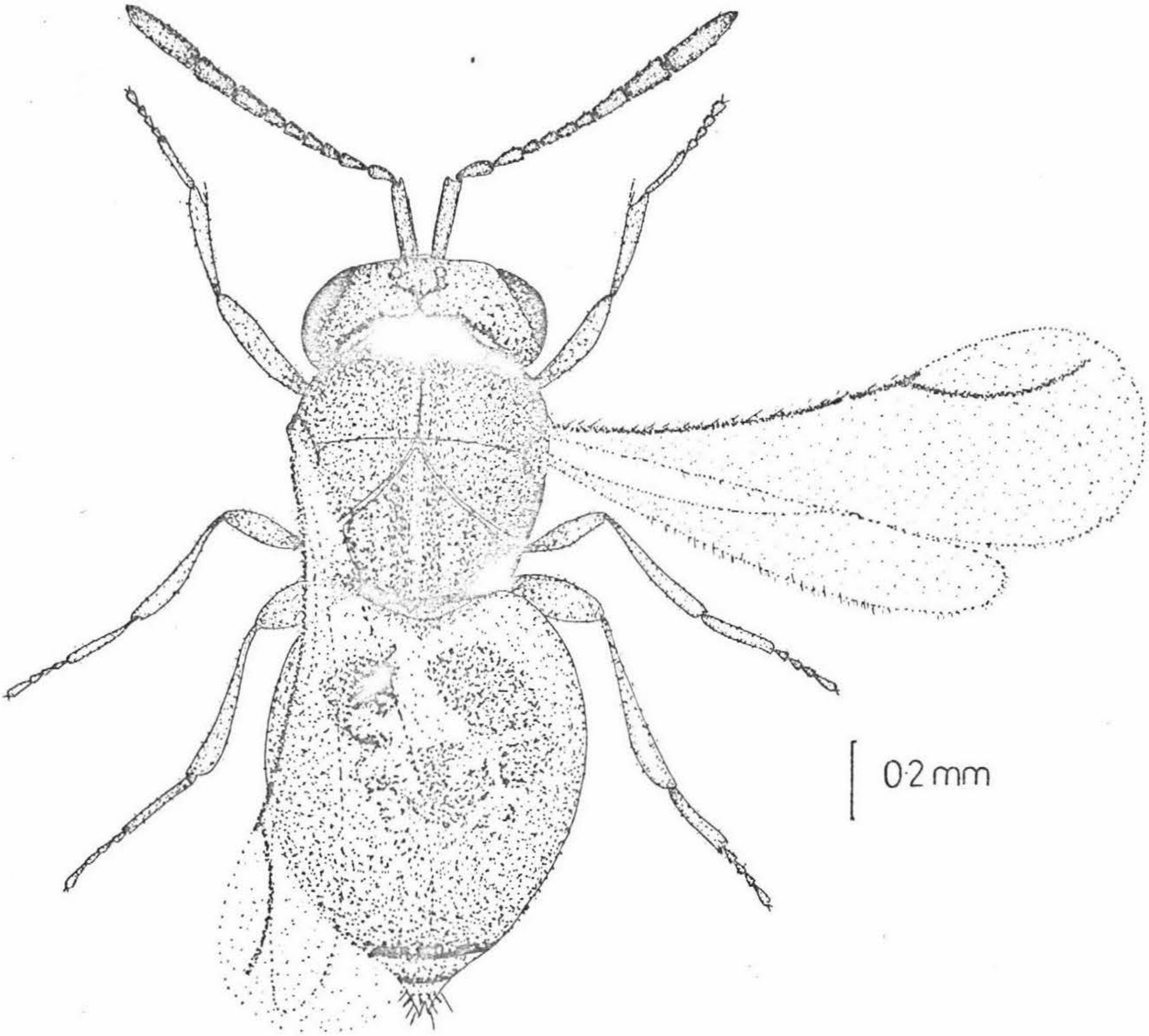


Fig.24 Adult female ceraphronid parasite of *P.radicis* .

of the parasite (Figure25). Pupation takes place within the gall.

All stages of development were observed in the relatively short period of P.radicis pupation (approximately 6 weeks). A number of galls dissected in the sample had already been vacated by the parasite. Only one pupa was observed in a gall at any other time of the year (over 1000 galls dissected). No adults were collected in the Malaise traps throughout the year but a number (ca.30) were extracted from soil samples (SectionII.9) two weeks after the emergence of P.radicis.

Oviposition was not observed in either field or laboratory conditions. Females when placed in the presence of galls ran over the galls, 'tapping' them repeatedly with their antennae. The ovipositor is retractile, pointed and well sclerotised. Parasite pupae removed from the galls rapidly developed into imagos. Newly emerged famales, when dissected, did not contain eggs.

The degree of parasitism of the pupal stage of P.radicis effected by the ceraphronid is approximately 20%. The relationship between parasite and pupal density is illustrated in Figure27 (Appendix Hyper-parasitism did occur; however, the greatest observed number of parasite pupae per host was only two (approximately 5% of the parasites observed resulted from hyper-parasitism).

Discussion

The parasite was tentatively identified as a member of the Ceraphronidae (superfamily Proctotrapoidea : Hymenoptera) by virtue of its wing venation and the presence of two tibial spurs on the foreleg (Figure 24)

Riek (1970) states Ceraphronidae as the only family of the Apocrita with two fore-tibial spurs. The identification was confirmed by Valentine (pers. comm.); however, further classification was not possible. Very little is known about the family. They are often collected in litter samples and have been reared from a number of Lepidoptera, Coleoptera,

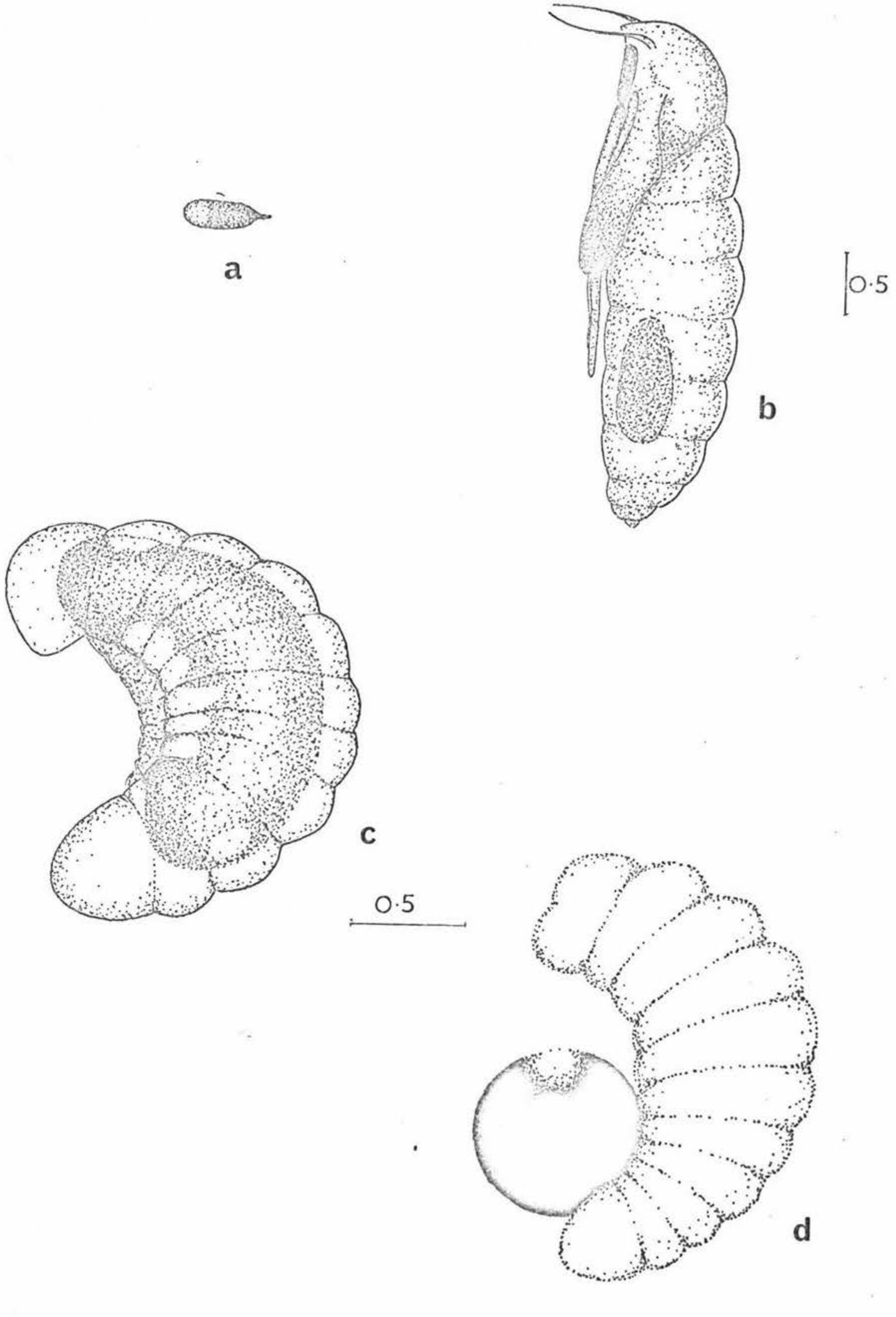


Fig.25 The developmental stages of the ceraphronid parasite ;
a) the egg, b) an early larva on a pupa of P. radicis,
c) mature larva and d) prepupa with shiny meconial pellet.

Diptera, Hymenoptera and Homoptera. However, exact relationships have only been determined in a few instances (Clausen, 1962; Valentine, pers. comm., 1972).

Haviland (1921) studied the bionomics and development of two ceraphronids (*Lygocerus testaccimanus* and *L.cameroni*). These forms are parasites of the larvae and pupae of certain *Aphidius* which are themselves internal parasites of various aphids. Spencer (1936) observed a similar biology for *L.niger*. Kamal (1939) reared the ceraphronids *Conostigmus zaglouli* and *C.timberlahei* from eggs laid within the puparium of a syriphid. It would appear then that most, if not all, ceraphronids develop externally to their host but within a cocoon, puparium, or dead body of a primary host. The ceraphronids can possibly only develop on soft, thinly cuticled, sedentary larvae or pupae. The parasite of *P.radicis* follows this propensity

Details of the full extent of the parasite's life cycle remain vague. As a complete egg-to-adult cycle was observed in summer it would appear that the parasite spends an extended, over-wintering stage as an adult, probably in the soil or litter. It is possible that it parasitises other insects but in view of the specialised nature of the attack on *P.radicis* this would seem unlikely. The adult is armed with a pair of large mandibles and probably feeds during its sojourn in the soil.

Early emerging adults may oviposit immediately and add to the degree of parasitism of *P.radicis* for that season; however, there was no evidence found to support this.

As all the eggs found were laid on the anterior region of the pupa, it is probable that oviposition takes place through the thinly protected exit-hole. The larvae, found attached to the host's pupal abdomen, must migrate from the original oviposition site. The larva appears to pierce the host's cuticle and feed by suction, gradually emptying the pupa.

The host pupa, normally capable of some movement, appears to be paralysed at an early stage of the parasitism. The host, although paling, does not show any sign of decay. The emergence of the parasite is presumed to be via the exit hole already prepared by the host larva.

Up to five eggs were found on some host pupae; however the competition between parasites in this situation appeared to be severe. In the double parasitism observed both were females and one was only half the size of the other (1 and 2 mm length respectively). As the parasite is approximately the same size as the host it is probably that successful hyper-parasitism can only be achieved on the female pupae of P.radicis as these are somewhat larger than the males.

The parasite appears to operate in a density-dependent manner (Figure27), although the sample is too small to determine definite trends, especially in the percentage parasitism at different pupal densities. The peak of parasitism in this sample comes at about 20 pupae/sample.

There is some difficulty in interpreting trends in the dynamics of a parasite population from one small sample. The results recorded are largely dependent on the dynamics of the preceding generation of the host. In this instance the parasite is also isolated from the host for a long period of time and any apparent density-dependent action may result from the effects of dispersion during the isolation.

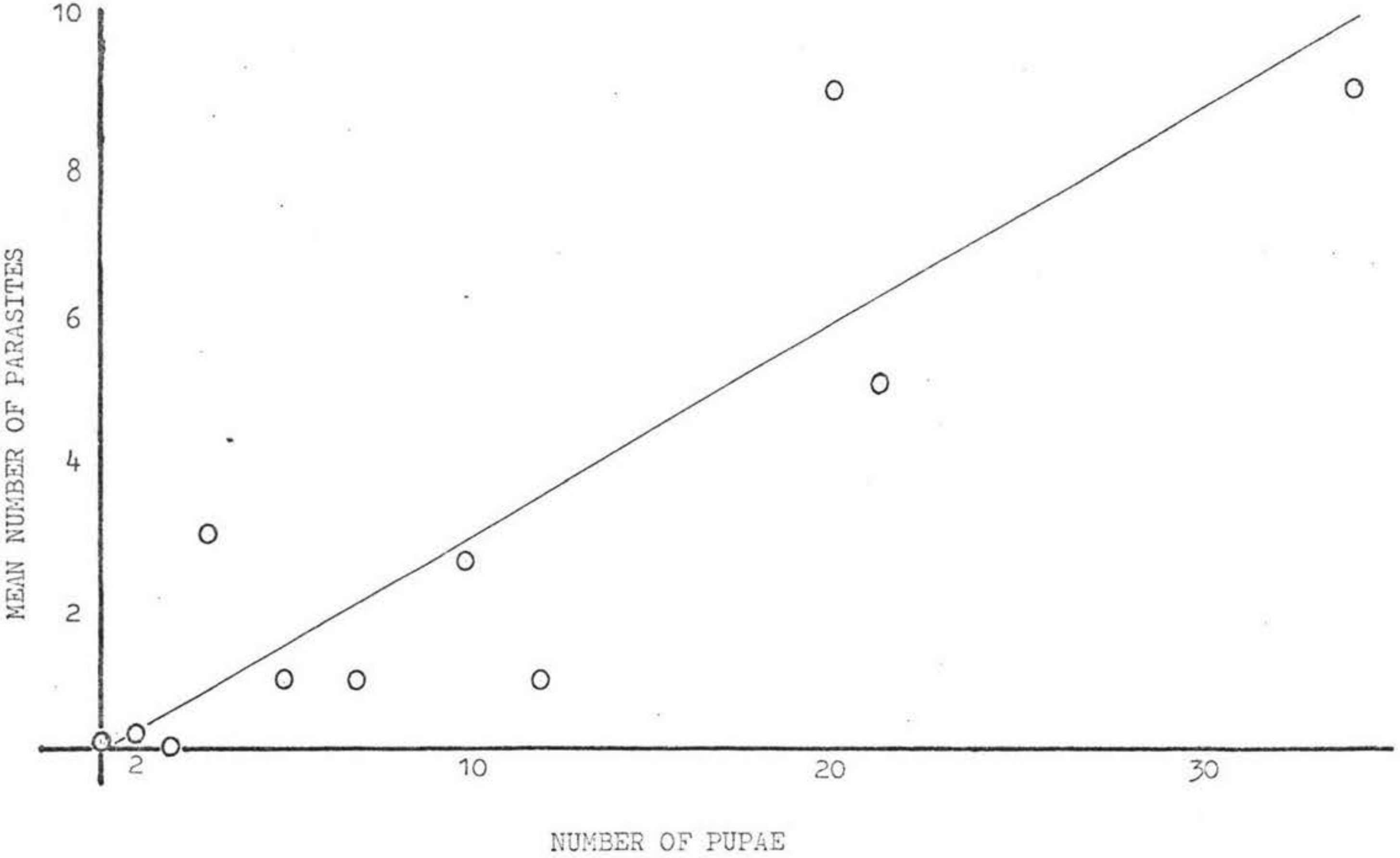


Fig. 27 The relationship between pupal and parasite numbers. (line by inspection)

A Life Table for P.radicis

A life table is a convenient method of collating data collected from various observations of the dynamics of a population: in this instance the observations of preceding sections. The reliability of the table is limited in that it only contains data from one generation. Typically life tables are built up over a number of generations and used for the prediction of future generations and the identification of 'key factors' (Morris, 1959) which influence the population.

Methods

Although only one generation is tabulated, an expanded version of Varley & Gradwell's (1960) population budget is used. For the purposes of the life table the population was considered stable, i.e. the generation studied was considered to have arisen from a preceding generation experiencing an identical total mortality. This allowed an assessment of the initial egg production (numbers entering the generation) from the results of the generation studied.

The average mortalities of the second and third larval instars and pupae were obtained from Section II.9. The maximum total fecundity was obtained from the results of female dissections in Section II.4, and counts of pupae per root weight were obtained from Section II.5 .

Adult mortality was considered negligible as their biological function is probably completed soon after emergence. Pupal counts represented the only quantitative data for the population entering a given stage; all calculations are based on, and relative to, this measurement. The initial calculation is adjusted to 1000 by the multiplication of which represents g weight of root needed to support this population.

Results

The relevant results from previous sections are tabulated, after the necessary conversions (Appendix 14), in Table XXI

The mortality during the life cycle of P.radicis is illustrated graphically in Fig.28.

Discussion

For convenience the conversions within the life table were based on a sex ratio of 1:1. Laboratory rearings (Section II.4) showed an actual sex ratio of 0.89 (male:female), i.e. egg production may in fact be slightly higher than predicted.

The survivorship curve of P.radicis is typical of type IV of Slobodkin's (1962) basic survivorship curves. It shows that mortality is greatest in the early unprotected stages of the life cycle. Mortality decreases while the larvae are protected within the gall but increases when the emergence exit is cut.

The mortality of the egg and first larval stages cannot be separated using the data obtained. However, both stages probably suffer from similar predation effected by eutytopic soil arthropods and nematodes. As the first larval instar has a longer duration than the egg stage, a greater degree of predation undoubtedly occurs during this stage.

Once within the gall, larval mortality drops dramatically and the larva enjoys a period of undisturbed growth and development. The exit hole, however, decreases the security of the gall and probably allows the entry of the ceraphronid parasite, predators and pathogens. The pupae represent a low point in the density of the insect population.
A comparatively small number of deaths (c.f. first instar mortality) could markedly affect the dynamics of the following generation. However, the parasitism encountered is not severe and it appears that the non-specific predation of first instar larvae is largely responsible for determining the

STAGE and probable mortality factor	Nos./11.05 g root	Log no.	% mortality in stage	K
MAXIMUM POTENTIAL NATALITY	$116 \times \frac{1.56}{2} \times 11.05$			
	= 1000	3.000		
K_0 (predation competition?)			97.3	1.569
SECOND LARVAL INSTAR	27	1.431		
K_1 (predation?)			11.29	0.051
THIRD LARVAL INSTAR	24	1.380		
K_2 (predation - fungus)			6.79	0.038
PUPAE	22	1.342		
K_3 (parasitism - other)			22.34	0.112
ADULT	17	1.230		
K_4		0		
ADULTS REPRODUCING	17/2 = 8.5* 9.3**	0.929		

TABLE XXI THE LIFE TABLE FOR P.radicis

(total generation 'mortality' K = 2.071)

* Number of reproducing females assuming a sex ratio of 1.0.

** Number of reproducing females applying observed sex ratio of 0.89 (male:female).

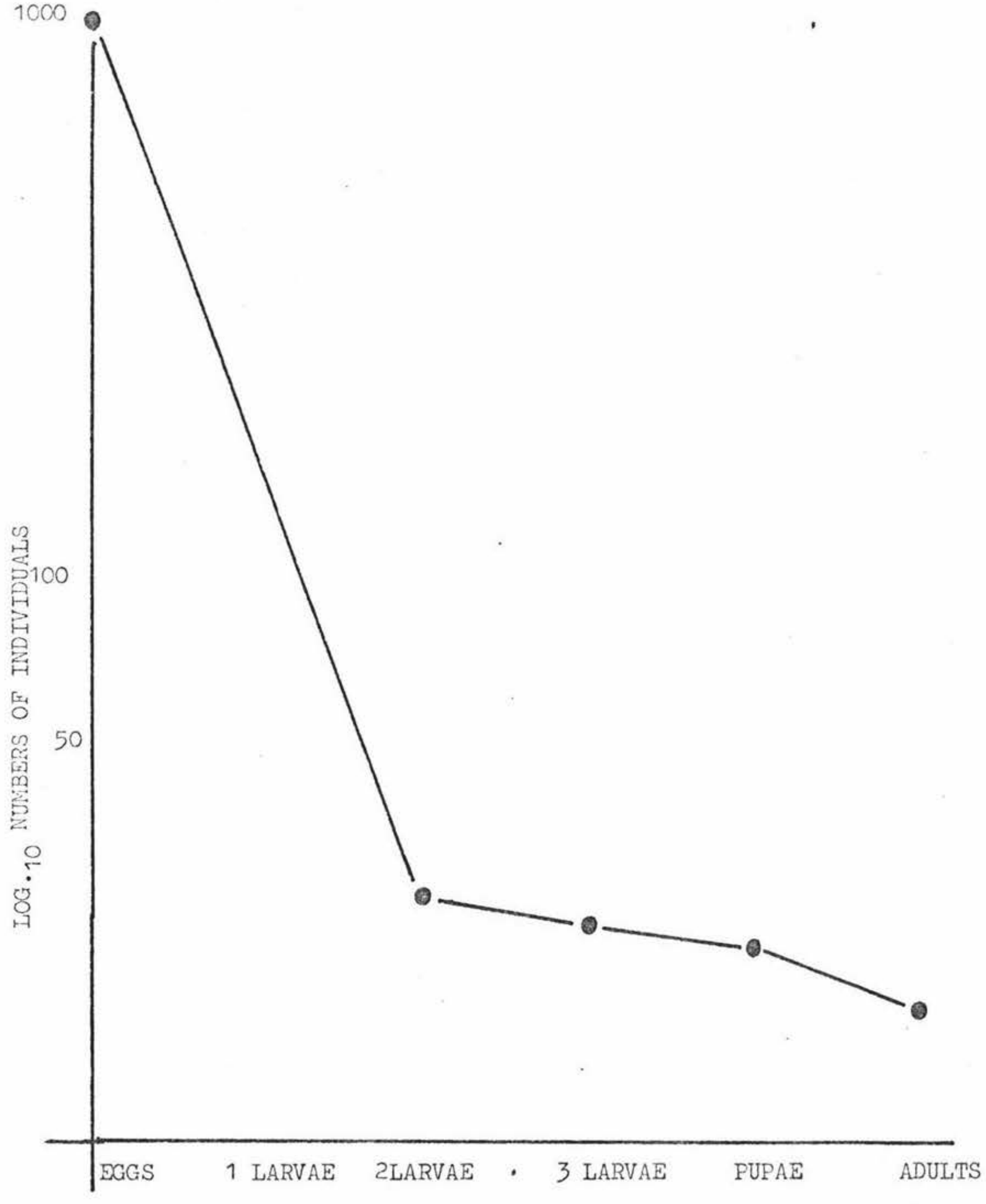


Fig.28 The survivorship curve for 1 generation of P. radicis.
(An initial correction of 11·05 produces a starting population of 1000.)

dynamics of the population. Such a large non-specific predatory complex probably imparts a degree of stability to the mortality of first instar larvae and the generation-to-generation fluctuation in numbers would not show the violent oscillation common to the two-member intersections of Nicholson (1933) and Utida (1957).

THE INSECT-PLANT RELATIONSHIP

Introduction

The presence of insects feeding on plants is often taken for granted. Moreover, the elevation of certain insects to pest status, by man, often promotes the idea of insects feeding voraciously on the defenceless green bounty. Phytophagy, however, is comparatively rare within the class Insecta. Only 8 of the 29 insect orders seriously utilise living plants as a food source. The greater proportion of insect orders contain scavengers or microbial feeders (Southwood, 1973). Those orders that have made the transition to phytophagy have so exploited the niche that numerically they may outstrip insects in other orders. The full significance of this insect-plant association, with respect to the individuals involved, has only really become apparent in the last 20 years.

A number of problems must be overcome during the transition from saprophagy to phytophagy. Nutrition and shelter are probably the most significant hurdles. Saprophagous insects feed on a varied mixture of substrate and micro-organisms in a relatively sheltered, humid environment. Plants, however, are often nutritionally inadequate (Schoonhaven, 1969) and phytophagy usually requires simultaneous adaptations against water-loss and predation. Cuticular specialisation by the insect can often account for the latter two elements, while many insects overcome nutritional barriers by feeding on a range of suitably endowed plants. However, as a more specific relationship develops between the insect and the plant the insect becomes more dependent on the equity of the plant.

Monophagy may allow greater ecological specialisation but the insect diet becomes subjected to the variations in composition that result from the plant's own interaction with the environment. Externally feeding monophagous insects may overcome this problem by feeding on a number of plants or dispersing to another area. Internally feeding insects (gall midges, leaf miners, etc.) are usually only capable of this choice in the initial stages of their life cycle. They may be effectively protected from desiccation, and to some extent predation, but in the process they are confined to one host plant for the greater part of their growth and development.

The effect of the insect feeding on the plant varies with the form of the attack. The majority of leaf-feeding insects do little more than reduce the effective photosynthetic capacity of the plant. Only a prolonged or concerted attack will significantly alter the host's physiology (Varley & Gradwell, 1960). In some cases the 'pruning' may be advantageous to the tree (Jackson & Osborne, 1970). Plant piercing insects of the Hemiptera may effect a change in the host's physiology which belies the amount of material extracted. Miles (1965) regarded a number of compounds in the saliva of aphids which could directly modify the host cells' metabolism. Dixon (1971) showed that leaves of sycamore and lime, when attacked by a psylid, are darker in colour and contain more nitrogen at leaf-fall than normal leaves. Similar phenomena have been observed between many leaf miners and their hosts (Engelbrecht <u>et al</u>., 1969). For the insect the advantages of altering the host's metabolism may be twofold: it overcomes seasonal variation in the nutrient composition of the plant organ on which it is feeding, and in some cases it may prolong the longevity of the plant organ.

Gall insects, especially Cecidomyiidae and Cynipoidea, are capable of inducing highly specialised abnormal plant growths in the immediate neighbourhood of the feeding larvae. The induced gall provides both nutrient and shelter for the insect. The gall is usually located in or near a meristematic region, which is rich in metabolites. The continued meristematic activity of the gall tissues acts as a sink, drawing metabolites to it (Jankiewicz & Plich, 1970) and lessening any seasonal variation. The degree of cellular specialisation within the gall varies with the insect and host involved. Some galls consist merely of a callus of undifferentiated cortical cells while others incorporate anatomical complexities specific to the gall. The mechanism of gall formation which produces this remarkably specificity of structure is not yet elucidated. It is generally conceded that the stimulus for gall formation is chemical as the associations formed are highly specific and cannot be mimicked by simple mechanical injury (Mani, 1964).

Plant growth and development involves a number of different biochemical systems. It is almost certain that different insects differ as to which system, or which part of a system, they influence. Hemiptera, for example, are known to secrete polyphenol oxidase which allows the accumulation of indol-acetic acid about the feeding site which in turn promotes gall formation (Miles, 1969). Hymenoptera appear to utilise nucleic acids in gall formation (McCalla, 1962). Dieleman's (1962) work implies that Cecidomyiidae re-secrete the plant's own hormones to promote a wound reaction about the feeding larva. Although the galls may be anatomically very specific, Petel *et al*. (1960) found that for the gall of *Viteus vitifoli* no permanent change in the requirement for auxin and cytokinin, to maintain callus growth, was apparent.

The plant is not necessarily a passive recipient of an insect invasion. The success of weed control programmes utilising insects and the persistence of resistant plant strains illustrate the selective forces which mould insect-plant relationships. Gordon (1959) refers to 'an unending biochemical contest between metazoans and the food organism'. The contest may, however, evolve into a compromise or mutual tolerance. Plants utilise phenological, physical and chemical resources in defence against insect attack. Phenologically the plant which develops past a susceptible stage before insect attack will gain selection. Many plants have a thickened cuticle and epidermal trichomes and glandular hairs which deter or trap feeding insects (Levin, 1973). Barrays & Chapman (1970) believe cuticle thickness is particularly important in the defence against small insects.

Chemically plants may contain a large array of so-called secondary plant compounds (alkaloids, phenolics, glycosides, essential oils, saponins, etc.): these compounds do not appear to be necessary for normal plant growth and development and were, for some time, considered to be waste products of primary metabolism. Fraenkel (1953,1959,1969), Dethier (1954,1970), Thorsteinson (1960), Jerømy (1966) and others advance the view that these compounds have been evolved as defence mechanisms against browsers, pathogens and competitors. Some of these compounds have insecticidal properties (nicotine, derris, pyrethrum, etc.). However, most of the interaction between insects and these compounds appears to be sensory and may act as feeding deterrents rather than eliciting a direct toxicological effect. In some cases insects appear to have overcome any deterrent effect and use the compounds as a means of host identification or even developmental cues (Ellis et al., 1965; Vite & Pitman, 1965).

Quantitative variation in primary and secondary plant compounds is a common seasonal occurrence. That this in itself may have a direct ecological effect on insect parasites is examplified in the work of Fenny & Bostock (1968) and Van Emden (1966,1971). Qualitative changes, however, may also occur, especially in response to damage or pathogens. Often pathogen invasion leads to the accumulation of various aromatic compounds (flavonoids, anthocyanins, phenolic glycocides, etc.) which check the infestation (Farkas & Kiraley, 1962). There is a great volume of literature on the post-infectional increase of phenolic compounds in plants, attacked by bacteria and fungi (review Rubin & Artsikhovskay, 1963). Similar responses have been shown in plants attacked by insects (Hillis & Inoue, 1968; Thielges, 1968). Galls are noted for their abnormal accumulation of polyhydroxyphenols or their derivatives and may contain as much as 75% tannin on a dry weight basis (Newcombe, 1951). Such plant responses are interpreted (Miles, 1968) as would reactions which attempt to isolate the invasion of the pathogen or parasite.

The subject of allelochemics, the chemical interaction between species (Whittaker & Feeny, 1971), is rapidly become recognised as a basic component in plant and animal ecology. For insects, chemical interactions are often implicated in host selection and feeding responses (Dethier, 1970), developmental cues (Ellis et al., 1965) and defence (Brower et al., 1968). More intimate insect-plant associations are the result of two co-evolving systems in which small changes may have major ecological effects. The extent to which chemical warfare is evoked probably determines the stability of the association.

Host Specificity

The feeding habits of the insects within the phytophagous insect orders are thought to have diversified as a response to the differentiation of higher plants (Wigglesworth, 1968). However, in concomitance with the increasing complexity of plant morphology and physiology, and in the interests of ecological efficiency, many insects have become restricted to a limited number of plants (Schoonhoven, 1973). This restriction is interpreted, in the majority of instances, as an active selection by the insect involved (Dethier, 1954; Fraenkel, 1969).

It is generally accepted that the monophagous feeding habit developed from polyphagy (Huettel & Bush, 1972; Stride & Straatman, 1962). Monophagy, although restricting the nutritional selection of the insect, does allow for a greater ecological specialisation of the insect-plant assocation, e.g. by reducing interspecific competition and allowing the insect to adapt behaviour physiology, colouration etc. to a specific microhabitat. A plant gall represents a very specific microhabitat so it is not surprising to find a relatively high degree of host specificity among gall insects.

The Cecidomyiidae in fact do show a marked preference for certain plant groupings. They are largely restricted to the Angiospermae within which they show particular affinities for certain families (Salicaceae, Fagaceae, Compositae, Rosaceae - Mani, 1964). There was an early propensity to describe each new gall as being caused by a new insect species: however, a number of ecological studies have shown that certain gall midge species may have wide host ranges while others are singularly restricted. The common swede midge (Contarina nasturtii) was found to produce 4 different types of gall and to be capable of invading 25 different plant species.

Metrosideros robusta, the host plant of P.radicis, is a member of the family Myrtaceae. Only two other midge species have been recorded from the Myrtaceae (Felt, 1925). Of the 45 recorded species of Metrosideros (Balgooy, 1963), 11 are endemic to New Zealand. Within the study area, 2 species (M.diffusa and M.perforata) were closely associated with M.robusta. From other areas of the North Island, Cooper (1954,1958) has reported evidence for the hybridisation of M.robusta and M.excelsa (pohutukawa). He concluded that in localised coastal areas (Rangitoto I., Central Lakes) there was an "almost imperceptible introgression of one species into the other". His interpretation was based on foliar studies which showed that the most definitive characters of the two species were leaf width and stamen length. Against these two characters he plotted the more variable features of leaf length, internode length, and calyx tube length (TableXXII). Although the ranges of M.robusta and M.umbellata overlap no hybridisation between the two species has been recorded (Wardle, 1971).

Initial studies (Wyatt, 1963) indicated P.radicis was restricted in host range to M.robusta. Wardle (1971) observed that M.umbellata was not attacked by P.radicis. A survey of Metrosideros species closely associated with M.robusta was undertaken to confirm this host specificity.

Materials and Methods

Numerous root samples of the three Metrosideros species in the study area were investigated for the presence of P.radicis both in the field and the laboratory. Sections of rootlets (approximately 1 cm in length, 2 mm in diameter) of the three species were fixed in Carnoy's solution and then embedded in paraffin wax following normal dehydration and embedding procedures. Transverse sections (10-14 um) were cut by means of a microtome and mounted, unstained, in Canada balsam upon microscope slides.

TABLE XXII. SHOOT MEASUREMENTS OF <u>M.robusta</u>, <u>M.excelsa</u> AND <u>M.robusta</u> x <u>excelsa</u> (cm)
(Cooper 1954,1958 modified)

Character	*M.robusta*	hybrids	*M.excelsa*
Leaf width	1.0-1.5	1.5-2.0	2.0-3.0
Leaf length	0.6-1.2	1.2-2.5	2.5-3.5
Leaf length	3.1-5.9	6.0-6.9	7.0-9.4
Internode length	0.4-0.9	1.0-1.1	1.2-2.8
Calyx tube length	0.3-0.8	0.9-1.0	1.1-1.3

M.robusta, *M.excelsa* and their hybrids were studied for the presence of *P.radicis* in the areas of Taupo (Pohutukawa Bluff) and Rotorua (Mokoia Island). Both of these species have a relatively short flowering time and it was not possible to collect inflorescence material for hybrid analysis during this period. Hybrid analysis was achieved by averaging the measurements of leaf width and length of the first two pairs of mature 'three-year-old' leaves below the internode marking the penultimate year's growth (i.e. usually the oldest leaves on a branch system - see SectionI.3). Leaf samples were obtained by shooting down branches with a .22 or .303 rifle, climbing trees or collecting windfalls. Each tree sampled has assessed for *P.radicis* infestation by a thorough search of its root system.

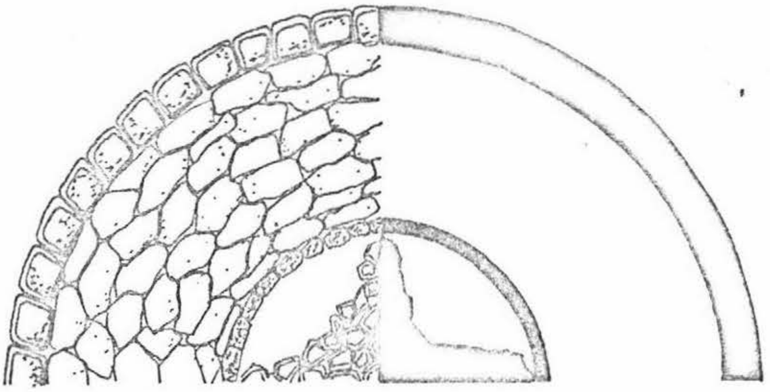
Results

The results from transverse sections of rootlets of *M.robusta*, *M.diffusa* and *M.perforata* are illustrated in Figure 29. A distinctive feature of the three species is the thickness of the epidermal cell wall. The epidermal cells of *M.diffusa* are uniformly thin walled. Those of *M.robusta* have the distal wall thickened while those of *M.perforata* were uniformly thickened and usually contained greenish unvacuolated cytoplasm.

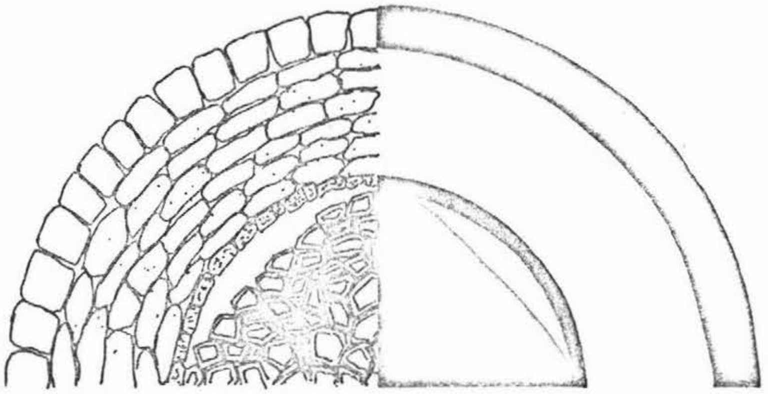
Galls formed by *P.radicis* were found only on the roots of *M.robusta*. No galls were found on either the hybrids of *M.robusta* x *excelsa* (Figure 30) or the closely attendant species *M.diffusa* and *M.perforata*.

Discussion

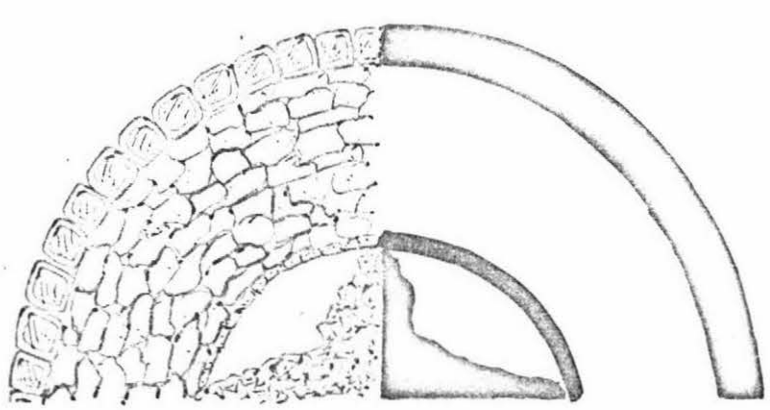
Anatomically the roots of the three species (*M.robusta*, *diffusa* and *perforata*) appear very similar. All have a tetrach vascular system although there is some variation in tissue ratios. Plant epidermal cells offer an initial barrier to an invading parasite and slight differences



robusta



diffusa



perforata

Fig.29 Diagrams of transverse sections showing differences between Metrosideros species.

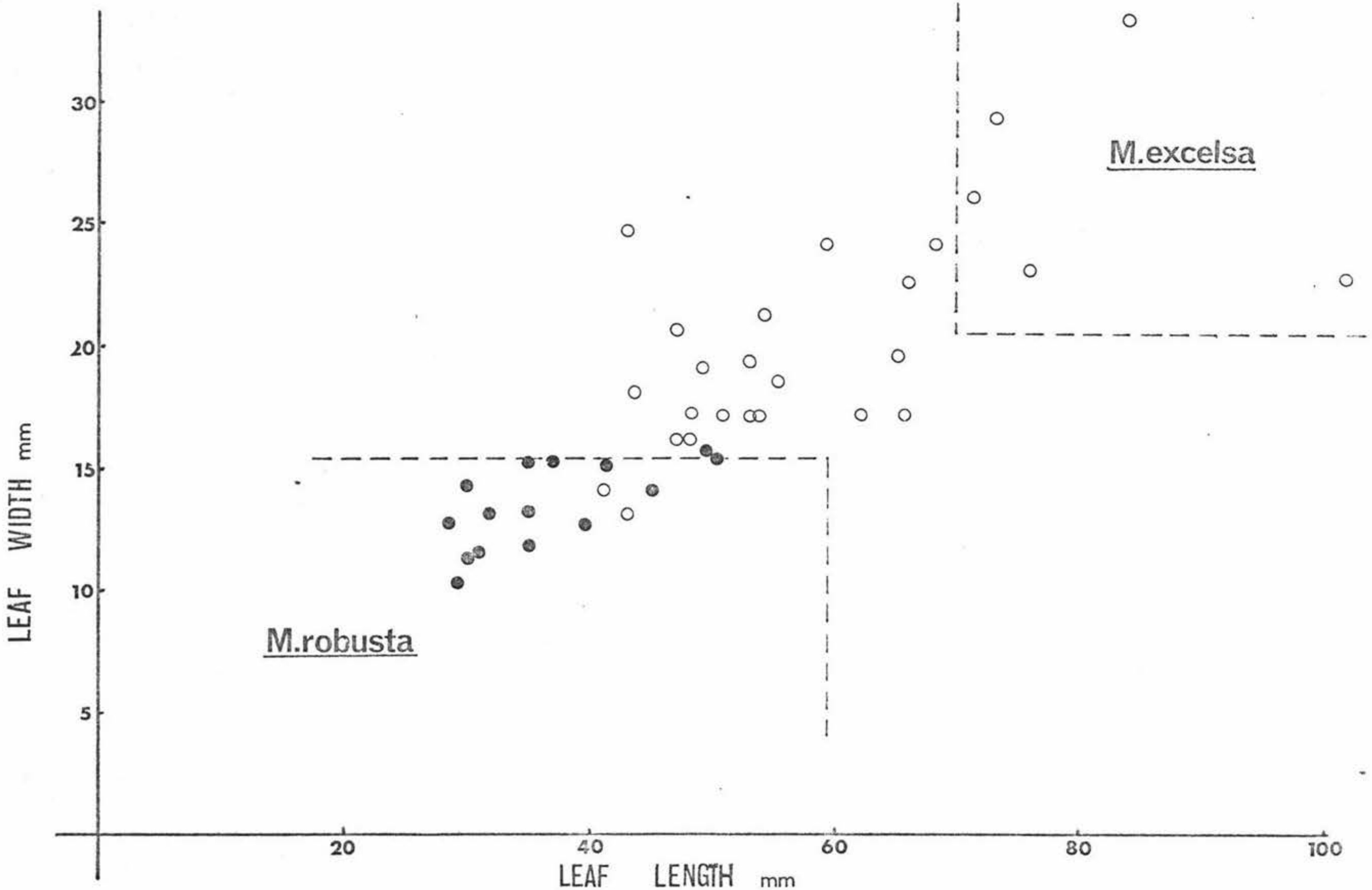


Fig.30 The infestation by P.radicis in the introgressive swarm of M.robusta x excelsa.
● infested ○ uninfested
-------- Cooper's (1954,1958) hybrid index

were recorded in these cells. The epidermal cells of *M.diffusa* were uniformly thin-walled. Those of *M.robusta* were thickened on the out wall while those of *M.perforata* were uniformly thickened and usually contained unvacuolated cytoplasm. On this evidence *M.diffusa*, rather than *M.robusta*, would appear to be the most susceptible to parasite entry. The preference of *P.radicis* for *M.robusta* is unlikely to be based on mechanical grounds.

From the results of the study of the hybrids of *M.robusta* and *M.excelsa*, it appears that *P.radicis* is restricted to unhybridised *M.robusta*. Even hybrids with a strong *M.robusta* element were not attacked. The specificity of *P.radicis* indicates that the insect can recognise its host plant from a milieu of closely related species. Although House (1969) has shown that quantitative differences in basic nutrients may be used for host selection in some insects, it is unlikely that significant differences in these compounds would occur between introgressive hybrids. So-called secondary plant compounds, however, may differ (Levin, 1971). Secondary plant compounds are used by many discriminatory insects for the initial recognition of host plants (Fraenkel, 1959,1969; Schoonhoven, 1969). These compounds may be detected at oviposition and/or during exploratory feeding by the larva. In many cases larvae are not confronted with a choice as the female oviposits explicitly on the host plant.

M.robusta has been shown to possess very few of the common secondary plant compounds (Table II). However, it is well endowed with phenolic compounds, the patterns of which are uniquely species-specific (Levin, 1971) in the majority of plants. Challice (1972) has shown that some phenolics tend to be inherited as simple dominant characters whereas others do not. There is a tendency for the phenolic patterns of hybrid plants to reflect a summation of the patterns of the parent plants (Glass, pers. comm., 1972). The introgressive hybrids of *M.robusta* and *M.excelsa* probably exhibit phenolic compounds common to both parent plants. Most selection by *P.radicis* is therefore probably based on attraction and repulsion. If only attractant compounds were utilised, *P.radicis* could be expected on *M.robusta* x *excelsa* hybrids. If only repellent compounds were used, *P.radicis* could be expected on a far greater range of plants, i.e. all plants not carrying the repellent compounds.

P.radicis, under laboratory conditions, was found to oviposit on or near root material (Section II.4). The first instar larvae are free-living but limited in their dispersal capabilities by their small size. The initial choice of a host plant is therefore made by the female while the immediate feeding site may be selected by the first larval instar. Both the adult and larva possess well developed antennae (Section I.4) For the larva, surrounded by 'acceptable' food, the selection of a feeding site must entail a very subtle discrimination, possibly incorporating tactile stimuli.

Gall structure

Plant galls, being conspicuous, attracted a lot of attention from early naturalists. Their structure has been documented by numerous observers from Malpighi to Mani (1964). Gall structure is dependent on a number of factors, not the least of which are the particular plant organ attacked and the identity of the parasite. Typically the gall structure is specific for a given host-parasite association; however, variation does occur with the time of attack and, in some cases, the sex of the parasite (Mani, 1964).

The specificity of the association strongly suggests the action of specific morphogenic hormones. (The very specificity of the reaction may be a prime factor in maintaining host preferences.) Gall formation involves the hypertrophy of the plant tissues either as a result of an increase in cell size or cell division. The change in cell structure is commonly accompanied by a change in cytology and gall cells generally have large nuclei and enriched cytoplasm (Mani, 1964). However, the minuteness of the gall insect and the small amounts of 'gall-inducing-principle' (McCalla, 1962) produced generally precludes chemical isolation. However, the morphogenetic action of a number of plant hormones and other compounds is reasonably well documented (Burstrom & Svensson, 1972; Steward & Krikorian, 1971) and it was considered that some insight into the nature of the insect-plant interaction might be gained from a study of gall structure.

Materials and Methods

Fresh galls of second instar larvae were fixed in Carnoy's Solution and embedded in paraffin wax as described in Section III.2. Transverse sections of the gall (10-14 um thick) were cut, mounted in Canada Balsam and compared under a binocular light microscope with similarly treated longitudinal sections of normal root.

Measurements were made of the length and breadth of cortical cells using an eyepiece micrometer. The cells measured were situated on imaginary lines perpendicular to the root epidermis. The measurements of 5 gall sections and 6 root sections were averaged.

As the number of cortical cells in the gall is somewhat greater than in the normal root, a comparison was achieved by dividing the cells into three regions (i.e. cortical cells adjacent to one epidermis, cells adjacent to the stele, and cells adjacent to the other epidermis). Each group contained approximately equal numbers of cells and the mean cell dimensions for each region were used in comparisons.

Results

The results of cortical cell measurements from gall and normal root tissue are expressed in Table XXIII and Figure 31. Plate 15 shows a typical gall section illustrating a distinct reduction in cell size in the vicinity of the larva.

Discussion

The results of cell counts and measurements shows that the amount of cortical tissue is increased to produce the gall. This increase is the product of an increase in cell number rather than a hypertrophy of individual cells. The gall, however, still retains the basic tissue pattern of the root, with a well defined stele. The similarity between root and gall tissue is also reflected by their similar dry weight percentages (Section III.4). Cell measurements suggest that cell expansion is inhibited at the site adjacent to the larva, while promotion of cell division may take place in the cortical tissue opposite the larva.

It is very difficult to identify functional mouthparts of phytophagous gall midge larvae (Dieleman, 1969). The lack of major cellular damage within the gall of P.radicis suggests that the larval mouthparts are small, possibly piercing. Dieleman (1969) suggests that

	Cell no.	Cortex ratio	Cell length	Cell breadth
Normal root	6:6	1:1	19.0	9.3
Gall	13.2:12.6	34.4:20.6	22.1:23.8	9.8:5.3
(distal:proximal) (to larva)		(1:0.6)	(1:1.07)	(1:0.54)

TABLE XXIII CORTICAL CELL MEASUREMENTS OF NORMAL AND GALLED ROOT -
A COMPARISON BETWEEN THE TWO CORTICAL REGIONS IN LONGITUDINAL SECTIONS
(dimensions in e.p.u.)

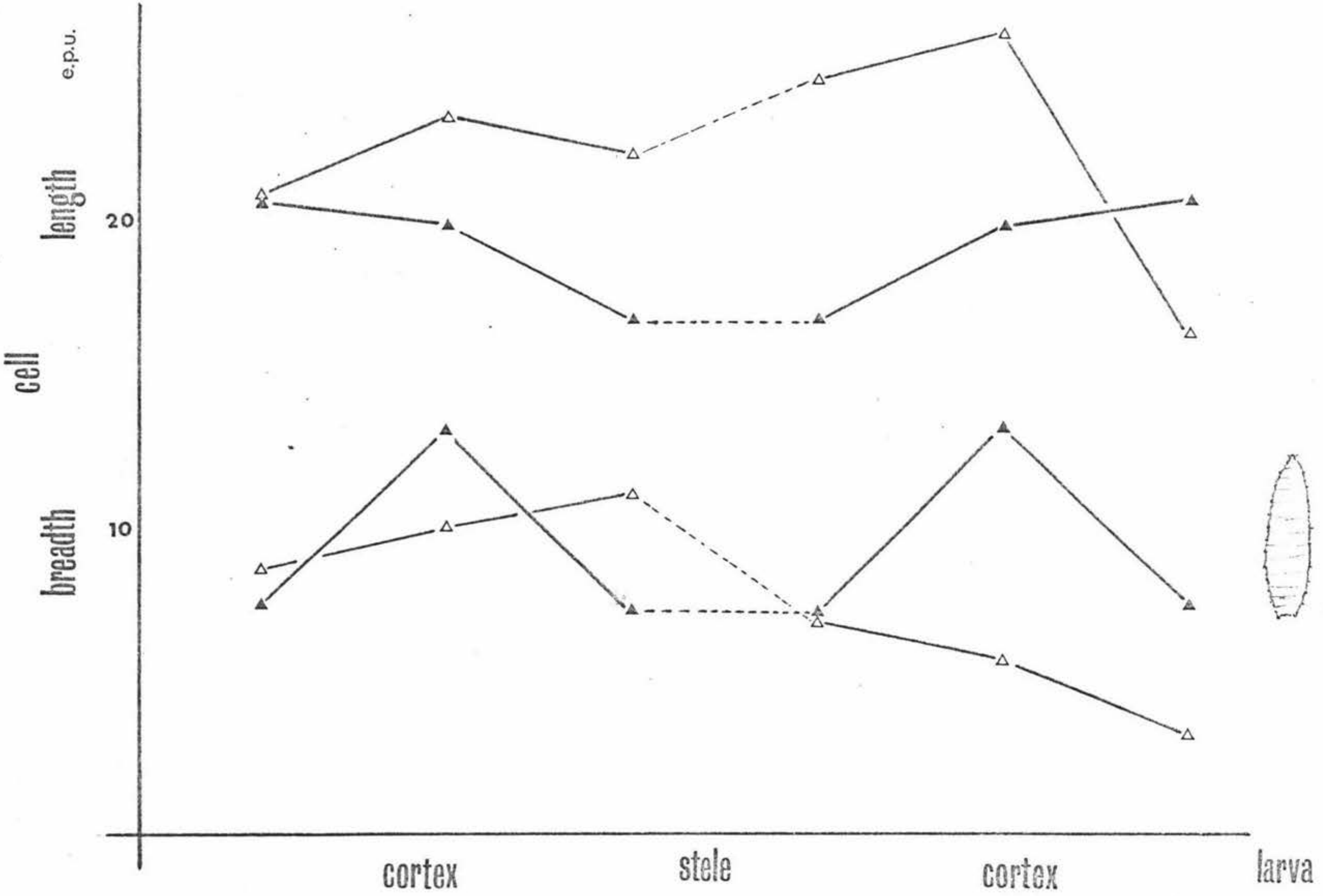


Fig.3l A comparison between the cortical cell dimensions of normal (▲) and galled (△) root.
(larvae are present only in the galled root)

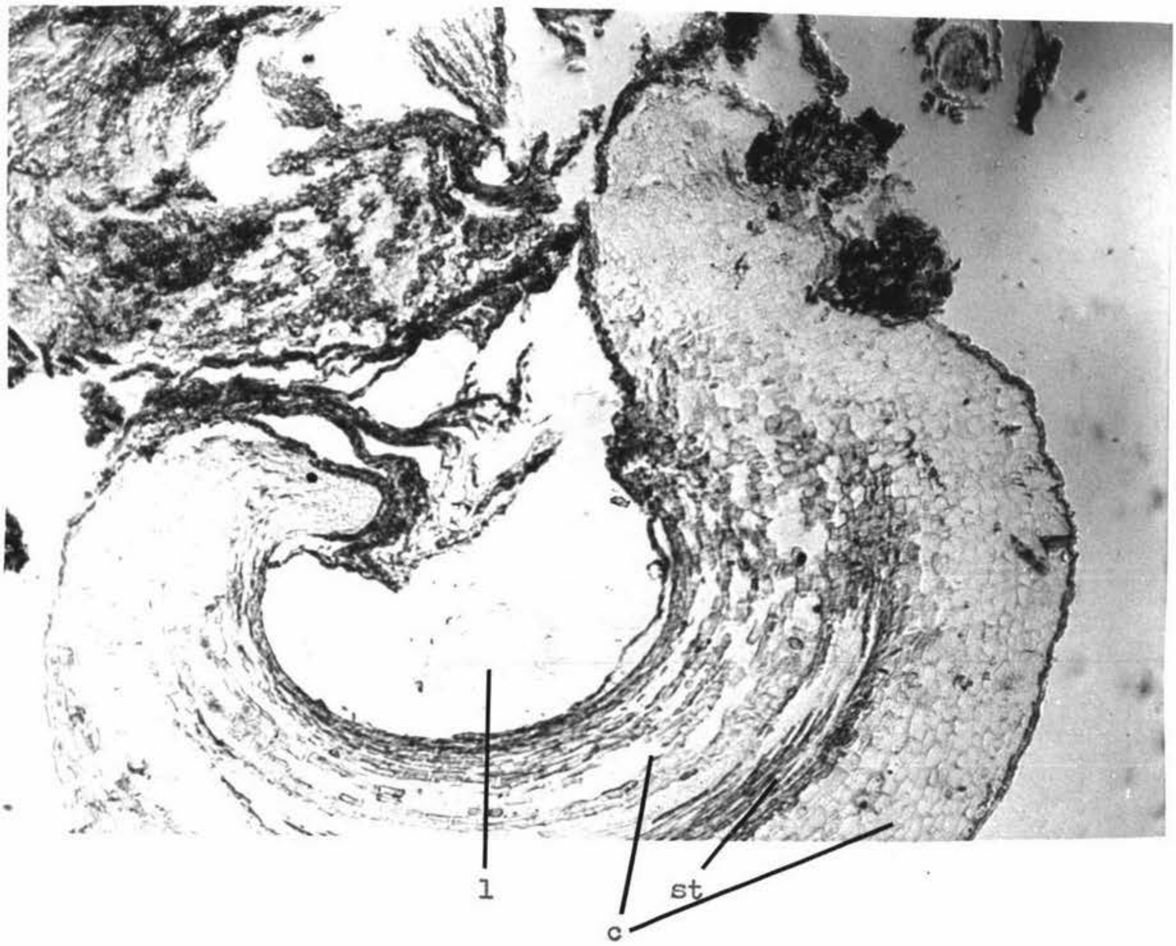


Plate 15 A section of a gall showing the position of the larva and the difference in the dimensions of cortical cells adjacent to, and removed from, the larva.
(larva as T.S. , root as L.S. - st = stele, c = cortex
l = larva)

cecids utilise the plant's own hormones to induce gall growth. However, as Clowes (1969) points out, the causal connections between plant hormones and root growth are still unknown.

Normal root growth requires all three classes of plant hormones (auxins, gibberellins and cytokinins) and the root meristem is a good source of hormones (Clowes, 1969). The action of exogenously supplied plant hormones on plant roots are listed in Table XXIV. The action of P.radicis appears to most closely mimic the effect of auxin. However, the number of compounds capable of stimulating or inhibiting plant growth is virtually limitless (Steward & Krikorian, 1971). Distinct plant inhibitors have been isolated from all plant organs (Hemberg, 1961) and a range of inhibitor-promoter compounds have been postulated for other cecidozoa (Miles, 1968; Bird, 1962; McCalla, 1962; Lecatsas, 1966).

It is interesting to note the levels of endogenous auxin in galls caused by different cecidogenic agents (Table XXV). The majority have higher than normal auxin levels. If auxin can be implicated in gall formation there appears no necessity to look further than Sheldrake & Northcote's (1968) finding for a source. They found that sterile animal and plant tissues, which had been killed by freezing and thawing, induced nodules of differentiated cells on previously undifferentiated callus of Phaseolus. They suggested that auxin was a normal product of autolysing cells and that dying cells were an important source of auxin in the plant. However, the mere presence of physical damage does not appear sufficient to promote distinctive gall formation (Mani, 1964). The continuous feeding of P.radicis, on the other hand, may be sufficient to induce the relatively simple gall on the roots of M.robusta.

The induced increase in root diameter may purely be a wound response. Roots of increased diameter are readily produced from primary roots after wounding or breakage (Section I.3). The switch from fine 'feeding-root' growth to 'runner-root' (Section I·3) growth is almost certainly hormonally

TABLE XXIV THE ACTION OF EXOGENOUS PLANT HORMONES ON ROOTS

Hormone		Reference
Auxins	1. Inhibit cell elongation, i.e. duration of cell elongation	
Indol acetic acid α-naphthylacetic acid	2. Reorientation of the direction of meritsem mitosis - tendency to radial rather than longitudinal growth - especially in the cortex. Cortex comes to occupy an increasing portion of the cross-section relative to the stele.	Burstrom & Svensson, 1972
Gibberellins GA_3	Little effect on roots or root segments.	"
Cytokinins Kinetin	Some swelling - growth inhibition. No change in mitotic polarity but some isodiametric swelling of cortical parenchyma. No great effect on cell elongation. Some other factor required for action.	" Torrey & Fosket, 1970

TABLE XXV · AUXIN LEVELS IN INVADED PLANT TISSUES

Organism	Auxin level relative to normal level	Reference
BACTERIA		
<u>Rhizobium</u>	above	*Thimann, 1936
(nodule bacterium)		*Link, 1937
		Link & Eggers, 1940
		Link <u>et</u> <u>al</u>., 1941
<u>Agrobacterium tumaefaciens</u>	above	*Klein & Link, 1955
		*De Ropp, 1951
(crown gall bacteria)		*Kulescha, 1949
VIRUS		
potato virus	below	*Jahnel, 1937
		*Lucas, 1939
		*Baumeister, 1951
FUNGI		
<u>Phytophthora</u>	above	*Dostal, 1950
<u>Cuscuta</u>	above	*Gustafson, 1946
<u>Omphalia</u>	below	*Sequira & Steves, 1954
NEMATODA		
<u>Meloidogyne</u>	above	Bird, 1962
		Balasubromanian & Rangaswami, 1962
INSECTA		
HEMIPTERA	above	Miles, 1968
Hymenoptera	-	Lecatsus, 1964
Cecidomyiidae	above?	Dieleman, 1969

* quoted in Hillman & Galston, 1961

controlled and probably induced by a build-up of auxin or some other wound hormone (traumatic acid, Wareing & Phillips, 1970). A gradient of this hormone may be sufficient to inhibit all expansion near the larva while promoting cell division further away. The continuous stimulation from the feeding larva is necessary for gall growth (Section II.3).

Gall Metabolism

An obvious effect of an insect's presence in a host is that it utilises the host's tissues to bring about its own growth. Gall insects induce meristematic action in the host's tissues which acts as a metabolic sink, drawing nutrients from surrounding plant tissues (Jankiewicz & Plich, 1970). For a rapidly developing insect the rate of nutrient draw-off may well be detrimental to the plant organ, or the plant as a whole.

The metabolic respiration of a gall should reflect the rate of nutrient turnover within it, and provide an estimate of the calorific cost of maintaining the gall. The respiration of normal roots and galls induced by P.radicis was investigated to determine the relative cost, to host, of maintaining P.radicis.

Materials and Methods

All plant material was obtained from the study area a day before experimentation and stored overnight at 4°C. Galls were stripped from roots and divided into white and brown galls. Undamaged roots, white galls and brown galls were placed in separate Warburg flasks with 2 ml distilled water. For the measurement of oxygen uptake, 2 pellets of KOH and 1 ml H_2O with a small filter-paper wick were placed in the centre cup of the flask. Oxygen uptake and CO_2 evolution were measured without KOH.

Oxygen uptake was measured from 20 to 0°C, decreasing in intervals of 5°C, using a Gilson Respirometer (constant temperature water bath). Readings at each temperature were taken at 10 minute intervals for one hour after a suitable equilibrium period. Each treatment was replicated at least three times and each run was accompanied by at least five 'blanks' (i.e. flasks containing only 2 ml H_2O and KOH solution) which served as controls. Although the Gilson Respirometer has a 'built-in' control flask, further controls were used for greater accuracy.

Treatments without KOH solution (O_2 + CO_2 exchange) were run at 20°C over 1 hour to determine respiratory quotients.

Results

The results of the respirometer experiments are recorded in TableXXVI. 'Fresh-weight' respiration of roots and galls is illustrated graphically in Figure 32.

Above 10°C normal root respiration was considerably higher than that of either second or third instar galls. Below 10°C all treatments showed a comparatively low rate of respiration.

At 20°C normal root tissue exhibits a respiratory quotient of 0.90 compared with 0.84 recorded for both gall tissue treatments.

Discussion

On a fresh-weight basis the rate of respiration of white galls approximates that of normal root at low temperatures. However, it drops to about a half of normal root respiration from 15 to 20°C. Respiration of brown galls (galls containing third instar larvae) is consistently lower than either white galls or normal root (Figure32). There was very little difference in percentage dry weight between the three tissues (23.3, 20.6, 22.0% roots, white galls, brown galls respectively) and conversion of respiratory rates to a dry weight basis only accentuates the curves (TableXXVI). The greater variation in the recordings of normal root respiration was due, in part, to the small weights of roots used in the experiments, as roots have a high volume to weight ratio. Also normal root treatments often consisted of a heterogenous mixture of differently aged tissue. There was some difficulty in obtaining sufficient root material to a uniform age without adversely 'pruning' samples.

The rate of increase of gall respiration is considerably less than that of normal root respiration. This suggests that the gall affords the

		Temp. °C					
		0	5	10	15	70	R.Q.
ROOT	Fresh wt	1.21	2.27	6.84	15.81	23.62	0.90
	Dry wt	5.19	9.76	29.32	57.14	98.38	
WHITE GALL	Fresh wt	1.74	2.76	4.26	7.33	10.75	0.84
	Dry wt	8.08	12.81	19.76	34.26	49.87	
BROWN GALL	Fresh wt	1.16	1.82	2.95	4.56	6.65	0.84
	Dry wt	5.39	8.45	13.68	21.25	30.80	

TABLE XXVI THE RESPIRATION RATES (ul 0_2/g/10 min.) AND RESPIRATORY QUOTIENTS OF GALL AND NORMAL ROOT TISSUE

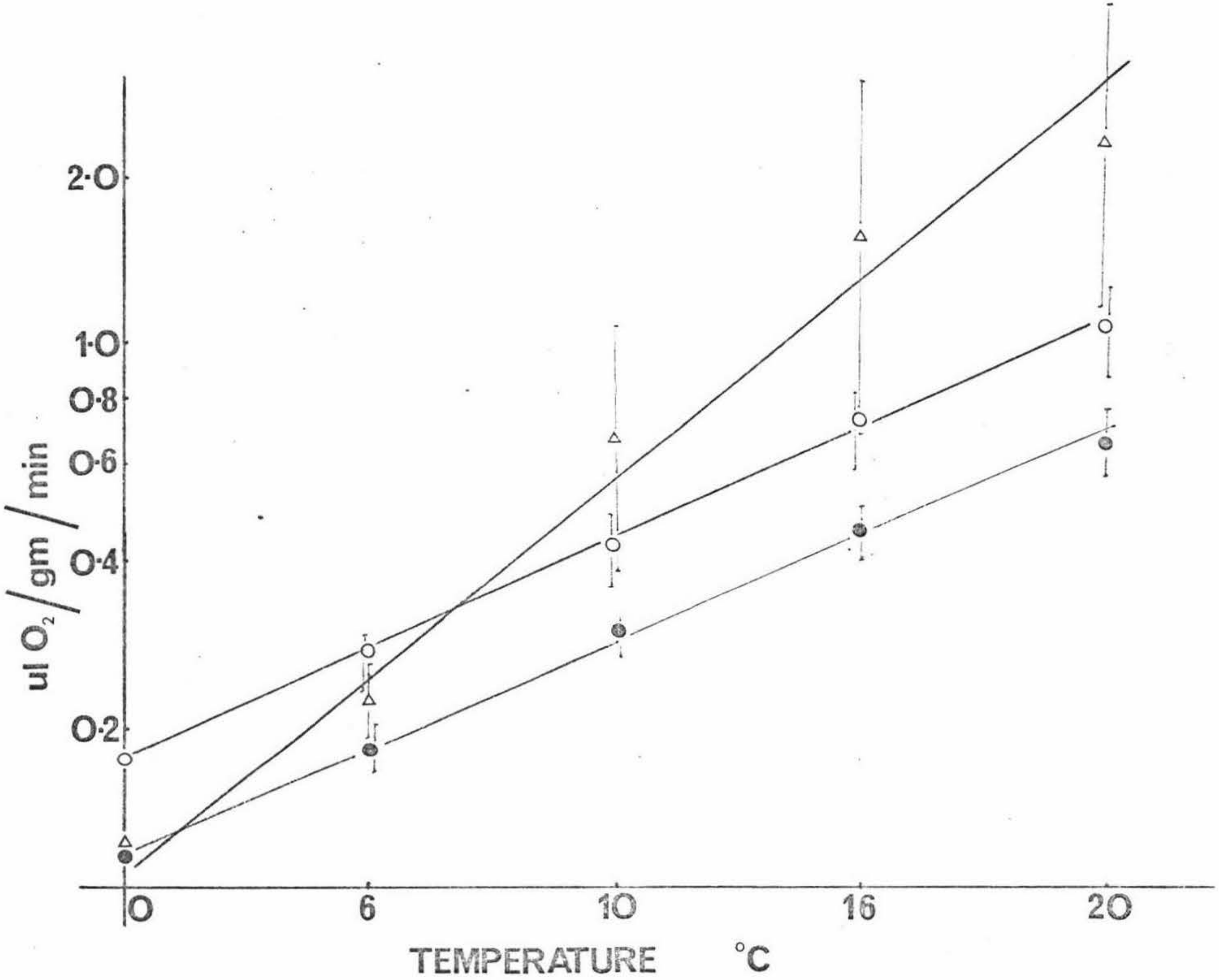


Fig. 32 The respiration rates of normal roots (△), galls of second instar larvae (○) and galls of third instar larvae (●) at temperatures from 0-20 C.

insect a more stable environment which is buffered to some extent against sudden fluctuations in temperature. The lower respiratory rate of old galls may in part be due to an unknown number of empty galls in the samples taken. However, it may also represent a lowering of the general metabolic activity of the gall and its inhabitant at the end of the insect's development.

The respiratory quotients (R.Q.) vary slightly for the different tissues (TableXXVI). If it is assumed that only carbohydrate and fat metabolism are used in respiration, normal roots (R.Q. 0.90) utilised 65.9% of O_2 consumed for carbohydrate metabolism. Both white and brown galls utilised about 48% of O_2 consumed for carbohydrate metabolism. However, a number of other metabolic pathways may have been in use, e.g. protein and alcohol catabolism (Southwood, 1966). Gall respiration is the result of two different metabolic systems, one insect and one plant, which may well account for the observed difference between gall and root R.Q.s.

The essential observation is that there is a qualitative difference between gall and root metabolism, and that for the greater part of the year, gall metabolism will be less than normal root metabolism as soil temperatures drop below 10°C for only four months of the year (Figure3) (SectionI.2)

Gall Chemistry - the Host's Defence

Plants are rarely entirely passive partners in an insect-plant association. The growth and development of an endogenously feeding insect is closely tied to the physiological conditions of the host plant and therefore susceptible to both quantitative and qualitative changes of the host's tissues. The presence of any degree of infection or lesion in a plant often leads to the accumulation of various phenolic compounds (Farkas & Kiraly, 1962). This accumulation has been construed as an active 'line of defence' against viruses, bacteria, fungi and insects (Levin, 1971). It has been suggested that the process was evolved by plants as a non-specific mechanism designed to contain or repel invading organisms (Miles, 1969).

Clasically, insect galls are associated with 'tannins', which are a heterogenous group of plant polyphenols (Brown, 1968). Their biological property lies in their 'astringency' (Bate-Smith, 1973), i.e. their ability to precipitate proteins. The accumulation of tannins in plant tissue is thought to precipitate the salivary enzymes of the invading organism and thus impede its development. Fenny (1967,1969,1970) showed that the accumulation of tannins in oak leaves significantly limited the growth of the feeding winter moth.

The quantity and quality of phenolic compounds in plant tissues can, in many instances, be correlated to the plant's resistance to intruders (Cruickshank & Perrin, 1964). The increased susceptibility to an attack may result from a lack (either constitutive or induced) of suitable phenolic compounds. The experiments of this section examined the adequacy of the defence of M.robusta in terms of the accumulation of phenolics resulting from an attack by P.radicis.

Materials and Methods

All plant material was obtained from the study area and transported to the laboratory in plastic bags. The time taken for the journey was approximately one hour. Material was stored at -10°C until treated.

A comparative analysis was made of phenolic quality, total phenol and leucoanthocyanin content, and 'relative-astringency' (Bate-Smith, 1973) for normal and galled root material. Relative astringency (a measure of the precipitation of proteins) was considered to be of greater ecological significance than a measure of total phenols.

For all analyses only young white roots (i.e. roots typically attacked by P.radicis) and the galls of second instar larvae, with larvae removed, were used. All three tests were applied to aliquots of similarly treated stock solutions. Galls were stripped from roots, dissected, and the larvae were removed. As wounding of plant material is followed by the accumulation of phenolic compounds (Sheldrake & Northcote, 1968), each gall, immediately after dissection, was plunged into hot 70% ethanol (approximately 50°C) to prevent enzymatic degradation. Roots were cut into sections (approximately 1 cm long) and treated as described above. The alcohol was then decanted from the plant material and retained separately. The plant material was weighed and then ground to a fine slurry with a pestle and mortar. The alcohol was added and the resulting suspension was refluxed (to gain an exhaustive extraction) for 48 hours in a soxhlet apparatus with approximately 100 ml 70% ethanol.

The resulting solution was evaporated to dryness at 40°C under reduced pressure and then taken up in 5 ml of distilled water. This served as a stock solution and was stored at 3°C (in a domestic refrigerator) until required. Each stock solution was adequate, when further diluted, for each test (total phenol, total leucoanthocyanins, and relative astringency) to be repeated at least three times.

Test for total phenols (TP)

Total phenol analysis was carried out by a slightly modified method of Swain & Hillis (1959).

0.5 ml Folin-Ciocalteau solution (diluted 1:1 H_2O) was added to 1 ml of the solution to be tested. After 3 minutes 2 ml of 20% Na_2CO_3 was added and the tube shaken. 2.5 ml of distilled water was added and the blue colouration was allowed to develop for 1 hour. The optical density of the resulting solution was read at 700 mu with an Eel Spectrometer using a water-reagent blank. A standard curve of 0, 10, 20, 50 ug/litre of phenol (B.D.H. reagent grade) was produced using the same procedure.

Test for leucoanthocyanins (LA)

The test for leucoanthocyanins was identical to that of Swain & Hills, 1959. However, a standard curve was obtained from serial dilutions of 'gall' solution and the results of normal root LA content were expressed as relative to the LA content of the gall solution.

Test for relative astringency (RA)

The test for relative astringency essentially followed that of Bate-Smith (1973). However, both human and frog haemoglobin was used. Although obvious colour changes occurred in treated solutions, their optical density could not be ascertained as they remained colloidal. Centrifugation at 6000 rpm for 10 minutes failed to clear the solutions. Instead, relative astringency was ranked in accordance with the amount of haemoglobin precipitated by the test solution as compared with suitable dilutions of 0.1% tannic acid solution. Precipitation was gauged by inspection as the precipitation of haemoglobin was quite abrupt and an estimation within 0.2 mg/ml tannic acid equivalents could easily be determined.

Qualitative phenolic pattern

20 ul of stock solutions were run by two-dimensional descending chromatography as in Section II.2. Chromatographs were developed under U.V. (with NH_3) and ferric chloride: potassium ferricyanide solution (equal volumes of 1% solution).

Results

The results of total phenol, leucoanthocyanin and relative astringency analyses are expressed in Table XXVII (data and standard curves in Appendix 18). Root tissue from galls shows a marked increase in leucoanthocyanin content and relative astringency when compared with normal root tissue.

No difference could be detected in the qualitative pattern of phenolics between normal and galled roots: the patterns obtained were replicates of that in Figure 12.

Discussion

The phenolic content of galls is consistently higher than that of normal roots. However, the results of the Swain-Hillis test for total phenols should be accepted with some reservation as other reducing substances may react with Folin-Ciocalteau reagent (Keith _et_ _al_., 1958). The standard curve for leucoanthocyanins is not strictly linear; however, dilutions of test solutions were arranged so that optical densities were approximately equal for both normal root and gall solutions.

Leucoanthocyanins and relative astringency appear to rise at the expense of other phenolics as total phenol content does not rise greatly and no qualitative change in phenolic pattern could be detected.
The levels attained in this induced resistance by the plant are not great when compared with the 10-20-fold increases found by Sequeira (1967), Hughes and Swain, 1960, and Kuc (1966) during fungal infections of various

SAMPLE	TOTAL PHENOL as phenol equivalents as % fresh weight	LEUCOANTHOCYANINS as ROOT relative to GALL	RELATIVE ASTRINGENCY as tannic acid equivalents as % fresh weight
ROOT	5.5×10^{-4}	23	4.1
	3.82×10^{-4}	22	3.35
	2.28×10^{-4}	38	4.70
GALL	6.35×10^{-4}	100	9.50
	7.85×10^{-4}		9.45
	4.15×10^{-4}		7.35
ROOT:GALL	1:1.66	1:3.6	1:2.13

TABLE XXVII THE PHENOLIC CONTENT OF NORMAL ROOT TISSUE AND GALL TISSUE INDUCED BY <u>P.radicis</u>

other plants. The lack of a strong response to invasion may account for the susceptibility of M.robusta to P.radicis, and to a number of other insects (Table III).

For gall insects, the accumulation of phenolics may not pose a serious problem. Nierenstein (1930) showed that Cynipid larvae (gall wasps) secrete tannase to hydrolyse tannins to gallic acid and thus prevent the precipitation of its salivary enzymes. P.radicis may be incorporating a similar defence as the difference in LA (tannin precursors) content between roots and galls is not morrored by a similar difference in relative astringency. If LAs are converted to tannins, P.radicis may be effecting their hydrolysis.

The build-up in LAs may not necessarily be entirely detrimental to the insect. P.radicis may well capitalise on such an accumulation. Leucoanthocyanins have been shown to act as synergists with auxin in promoting cell growth in various bioassays (Steward & Krikorian, 1972). LAs are capable of inducing cell division in carrot root phloems and have been shown to occur in many fruits and in the vicinity of developing embryos. The wound response of M.robusta to the invasion by P.radicis may well enhance the formation and maintenance of the induced gall. The accumulation of phenols may act as an adequate defence against secondary infection from less specialised soil fungi, nematodes, etc. No fungal entry into the root system as a result of gall formation was detected during the study.

The source of defensive phenolic compounds is not known. They may be synthesised at the site of infection or translocated to that site (Brown, 1964). From studies of Eucalytus spp., Hillis (1955,1956) concludes that leucoanthocyanins are synthesised in growing leaves from which they are later translocated, as glycosides, in the phloem. Hillis (1958) believes leucoanthocyanins polymerise, either alone or with other compounds, to form condensed tannins.

GENERAL CONCLUSION

Wyatt (1963) suggested that taxonomically *Proterodiplosis radicis* would be best regarded as a primitive member of the sub-tribe *Trifila* (Tribe Cecidomyiini). Felt (1925) tentatively places *Lopesia* Tavares and *Allodiplosis* Kieffer & Jorgensen, the genera most closely related to *Proterodiplosis*, in both the Cecidomyiini and the Porricondylariae tribes. There is little known about these two genera: *Lopesia* is recorded from Africa and *Allodiplosis* from South America (Felt, 1925). Both are probably too far removed from *P.radicis* to bear any close ecological similarity. Even within genera the diverse biologies of cecids preclude the recognition of general trends.

The taxonomic position of *P.radicis* is reflected by the insect's general biology. Although inducing a distinctive gall on the roots of rata, *P.radicis* has not left the moist primeval soil environment. The gall formed is a relatively simple fold-gall in which the insect is enclosed by the comparatively slow growth of the root meristem. Within the range of feeding habits exhibited by the Cecidomyiidae, *P.radicis* probably represents a transition stage between the soil-inhabiting omnivores and the truly arboreal phytophagous endoparasites. Within its forest soil environment *P.radicis* does not have to contend with the problems of desiccation and attachment typical of the arboreal habitat (Southwood, 1973). *P.radicis* shuns the drier soil regions which could create a water stress. Moreover a plant root system offers an ideal transitional medium. The root growth of temperate trees continues for a longer period than shoot growth (Kozlowski, 1972). The virtually continuous growth of the insect's food supply does not demand the close phenological synchrony often encountered in life cycles of aerial gall midges (Johnson, 1963). Instead, the biology of *P.radicis* appears to be more closely synchronised with the weather.

The life cycles of cecids can be exceedingly complex and may involve larval and pupal paedogenesis and parthenogenesis (Harris, 1968). P.radicis, however, exhbits no unusual reproductive phases. The life cycle basically has a duration of one year. The first larval instar may be prolonged, by facultative diapause, in adverse weather conditions or through competition for a feeding site. A small percentage of the population enters diapause in the third larval instar. Diapause is relatively rare in the New Zealand insect fauna (Dumbleton, 1967); however, it is of obvious advantage in the life cycle of P.radicis where the fragile, ephemeral, adults represent the sole reproductive stage. The degree of diapause exhibited is small in comparison with that recorded for other cecids (Johnson, 1963; Hedlin, 1964) and in no way detracts from Dumbleton's hypothesis of relatively warm Pleistocene climates in New Zealand. A small degree of diapause in the population of P.radicis could be viewed as a prudent safeguard against untimely inclement weather. In this instance diapause could hardly be considered necessary for phenological synchronisation with the host.

Adult gall-midges are extremely delicate insects. Typically they do not feed and represent the reproductive and dispersive stage in the life cycle. Cecids are not strong fliers and their small size allows them to be readily dispersed by the wind. While this could be of benefit in some circumstances it could well be a disruptive influence during host or mate selection. P.radicis overcomes these disadvantages in a number of ways. Adult emergence coincides with the settled weather of February. The emerged females are found within the litter layer, a position which both resists dispersal and facilitates host detection. The male, although removed from the litter, is rarely any great distance from it and rests in sheltered pockets in the undergrowth. The cryptic behaviour of the adults necessitates some form of mate attraction.

Results and observations in this study indicate that the females utilise a pheromone to attract the males. Amongst the Cecidomyiidae, pheromones have previously only been implicated in the mating of the Hessian fly (Cartwright, 1922). However, the striking sexual dimorphism of antennae within the family (Felt, 1925) would indicate that pheromones are widely utilised.

The regulation and/or control of an endopterygote insect population usually involves negating factors in two distinct environments: larval and adult. P.radicis, however, could be considered to pass through three distinctly different environments during its life history. The adults are aerial; the eggs and first instar larvae inhabit the soil, while the second and third larval instars and the pupae occupy an endoparasitic niche within the host plant.

In considering the fragility of the adults the aerial environment would appear to be the most tenuous. The adults, however, are inherently short-lived and the population is adapted to suit their ephemeral existence. The emergence of adults is prolonged so that only an uncommonly long period of inclement weather would destroy the breeding population. Males, although shorter-lived than females and more exposed to climatic vagarities, may overcome any effects of increased mortality by polygamy. Moreover, male attraction and single matings by females ensures an efficient use of male gametes.

The egg and first instar larva of P.radicis are no less fragile than the adults. However, the egg-larval period is considerably longer than that of the adult and it is probably exposed to a higher degree of predation and competition. This stage represents the greatest density of the insect in its life cycle. The virtual immobility of the individuals within such a eutrophic environment must increase the chances of predation and the degree of both inter- and intra-specific competition. Not surprisingly, it is this stage of the life cycle which suffers the greatest mortality. The mortality is probably effected by a large number of eurytopic soil arthropods and at least one nematode species.

The development of P. radicis following the formation of the gall is relatively immune from the external influences of weather and predation. However, the host plant cannot be considered an entirely passive recipient of the entry of P. radicis. An apparent induced resistance involving the accumulation of phenolic compounds takes place. However, no larval mortality could be attributed to this accumulation and the induced reaction of the host is probably countered by the larva. Such exchanges are typical in the development and continuance of a host-parasite association (Miles, 1968).

The security of the gall is lost with the pre-emergence rupture of the larval cavity. The cutting of an exit hole late in the third larval instar exposes subsequent stages to predators, parasites and pathogens. The predators and pathogens involved are probably similar to those effecting the mortality of eggs and first instar larvae. The prepupal and pupal stadia, however, are attacked by a ceraphronid parasite. The Ceraphronidae is not a well-known family within the Hymenoptera (Valentine, pers. comm.) and the parsitism of P. radicis appears to be the first record of a cecid host. The degree of parasitism achieved, however, is small in comparison with the early larval mortality. The adult parasites were found within the litter in an environment similar to that of the early developmental stages of P. radicis. The host and the parasite may therefore be subjected to similar regulating forces. If so, the degree of parasitism may remain relatively constant, with a season of low larval mortality being mirrored by a similar parasite mortality. For P. radicis a high larval survival would subsequently be countered by a high pupal parasitism.

The dispersal of P.radicis (adults) appears to be heterogenous and it is not considered to have any major deleterious effect on the numbers in the population. The ovipositing female population is cryptic and stationary. The males, although more susceptible to dispersal because of their aerial habitat, may mate more than once so that some loss is not necessarily disruptive to the population. Within the area of the present study the host trees were sparsely distributed so that populations of P.radicis about individual trees were virtually separate entities. In such a situation immigration would be fortuitous and the majority of emigrating individuals could be considered lost to the population as a whole. Successful male emigrants would tend to widen the gene pool of the total population rather than provide a significant numeric increase.

Air-borne cecids tend to congregate on the leeward side of obstacles (Johnson, 1969) and an apparent dispersal effect is the skewed distribution of a population of P.radicis about a particular tree. The roots of the leeward sides of host trees tended to be more severely infested with galls than those of the windward sides. This pattern of distribution may well be a factor in limiting the severity of an attack on a tree.

The present study did not uncover any evidence of major numerical fluctuations in the population of P.radicis. The general stability of the soil environment probably imparts a degree of stability to the population. However, the soil environment is intimately associated with the debris from above-ground vegetation. The introduction of exotic herbivores into New Zealand's indigenous forest is undoubtedly influencing the balance of this ecosystem (McKelvey, 1965). Browsing affects the amount and the spatial and temporal pattern of debris distribution. In the short term, increased browsing of aerial vegetation may be beneficial to P.radicis, in that a less bountiful substrate would support fewer eurytopic predators. However, on the skeletal soils of the study area the root system of rata is very shallow. A decrease in the depth of the humus layer would eventually limit root extension, or the availability of the feeding sites of P.radicis. Defoliation also opens the forest canopy and subjects the forest floor, plant roots and larvae to desiccation.

A more subtle interaction between P.radicis and the browsers may exist. The opossums in the study area were observed to feed selectively on the young leaves of rata in winter, presumably when preferred foods were scarce. It is estimated that three successive years of feeding by the opossum is sufficient to effect tree mortality (Meads, pers. comm.). The leaves of rata, however, are rich in tannins (Betts, 1919), which are considered to be feeding repellants (Fenny & Bostock, 1968; Bate-Smith, 1973). If, as Hathaway (1968) postulates, leucoanthocyanins (tannin precursors) are translocated within plants, galls acting as 'sinks' for these phenolics may render the leaves more palatable to browsers; opossums and insects alike.

Evidence of a direct injurious effect caused by P.radicis on its host, M.robusta, is lacking. The fact that P.radicis is restricted to this host, and is present over its entire geographic range, suggests a long association of the two species, which in its original context probably developed to a mutual tolerance. The reaction of the tree to the continuous feeding of the larva is primarily a disturbance in the normal development of the root. The calorific cost to the tree in maintaining the gall is probably minimal as gall metabolism is generally less than that required for normal root respiration. The functional loss of the root tissue may be of more importance, especially for older trees. The galls have no root hairs and presumably play little part in the active uptake of nutrients. Although the loss of small rootlets from trees is common, there is some evidence that older trees are less proficient at root replacement (Kozlowski, 1971). Moreover, root growth is directly dependent on photosynthates from the shoot. Trees defoliated by browsers may be limited in their ability to regenerate roots (Redmond, 1959). Any loss of rootlets through browsing or non-regeneration, without a simultaneous decrease in the infestation by P.radicis, would lead to higher relative gall densities which would compound the problem.

There is a close developmental synchronisation between the insect larva and the gall. The resulting gall is essential for the normal development of the insect but there seems no necessity to implicate sophisticated 'gall-inducing principles' (Mani, 1964) in the production of the gall. The described associations and interactions between the accumulation of phenolic compounds, plant hormones and plant wound responses (Sheldrake & Northcote, 1968; Miles, 1968; Burstrom & Svensson, 1972) appear sufficient to produce the simple fold gall induced by P.radicis.

The future of either P.radicis or M.robusta is not bright. The activity of introduced browsers in the forest directly or indirectly destroys the thick, damp litter layer essential for the surface rooting of M.robusta. This loss directly limits the food source of P.radicis. M.robusta, on the other hand, appears to adapt reasonably well to this situation and may benefit from the loss of P.radicis. However, the regeneration of M.robusta is negligible. Terrestial seedlings are either eaten or die from desiccation while epiphytic seedlings are dependent on large emergent trees which are rapidly disappearing from lower montane forests.

It is clear from the study of Proterodiplosis radicis that this, and other phytophagous Cecidomyiidae, offer a unique opportunity to study an intimate association between a plant and an insect. Further study exploiting the knowledge and techniques thus far gained could benefit such diverse fields as plant morphology, plant pathology and crop protection.

Several aspects of the present study lend themselves to direct expansion. With little effort the mechanism of mate attraction could be established beyond doubt. The availability of plants from hybrid introgressions of the host offers a good opportunity for the investigation of host attraction. Obviously any subsequent study would benefit from the successful infestation of host plants propagated, and examinable, in the laboratory. Manageable infestations would afford an opportunity to observe, by isotope and analytical techniques, the cause and effect of an intimate insect-plant association.

SUMMARY

1. Phenology of the host was investigated. Although showing distinct periods of flowering and shoot growth, root growth appeared to be continuous.

2. The juvenile stages of P. radicis are described. Three larval instars are readily recognisable by virtue of their size, colour and presence or absence of certain cuticular structures.

3. The life cycle of P. radicis has a duration of one year. About 10% of the population may diapause as third instar larvae. Adult emergence is from mid-February to March.

4. The adult life span is short (2-4 days). Females have a total maximum fecundity of about 116. Chemical attraction appears to be utilised to assist mate selection.

5. Gall distribution is restricted in part by low soil moistures and soil depth. However, galls are usually present on all healthy fine rootlets. The population distribution tends to be shewed to the leeward side of the host tree.

6. Effective dispersal in areas of low host density is low. The geotactic behaviour of the female creates a heterogenous dispersal.

7. The main regulation of population numbers appears to take place during the first larval instar. This instar is free-living and may be prolonged, possibly through competition for a feeding site, and is vulnerable to predation by a large number of eurytopic soil animals and susceptible to desiccation.

8. The pupae of <u>P.radicis</u> are parasitised by a previously unrecorded ceraphronid parasite. The degree of paraitism is slight.

9. <u>P.radicis</u> is strongly host specific, attacking only unhybridised host trees. Both the host and the parasite appear mutually well adapted to each other and the parasite cannot be implicated in the reportedly widespread debility of the host.

APPENDIX I DATA FOR RATA PHENOLOGY - SHOOT GROWTH (cm)

Shoot no.	Date of measurement							
	21.1.72	30.1.72	4.2.72	11 2.72	18.2.72	3.3.72	24.3.72	5.5.72
1	6.5	7.0	7.0	7.3	7.5	9.0	13.0	9.5
2	6.5	7.7	8.6	11.0	11.0	13.0	13.0	9.5
3	6.9	6.9						
4	6.4	8.2	8.8	9.6	11.0	12.0	12.2	
5	8.4	11.0	11.0	12.0	12.8	13.2	12.6	12.6
6	5.3							
7	5.5	4.8	5.2	5.7	6.5	7.1		
8	8.1	10.5	11.3	12.5	12.5	12.5	13.0	12.9
9	9.5	11.5	12.2	12.5	13.1	13.0	13.5	
10	6.1	8.5	9.9	11.2	12.1	12.0	12.2	11.4
11	6.3							
12	8.3							
13	10.0	10.8	11.5	12.4	13.0	13.6	13.6	13.7
14	7.1							
15	4.5	4.8	4.3	4.9	5.4	8.0	10.5	11.2
16	8.5	9.0	9.4	9.4	9.4	9.4	9.0	9.5
17	8.6			8.5	8.7	8.7		7.0
18	3.5	5.2	6.4	6.5	7.5	9.2	9.8	9.0
19	6.0	7.0	7.0	7.0	7.0	7.0	6.5	
20	5.6	7.5	7.3	8.2	8.9	9.2	9.5	9.5
Av. weekly growth		1.13	1.12	0.74	0.8	0.64	0.29	0.1
No.of shoots aborted or missing		6	8	9	10	11	19	18
'biomass' accumulate		1.13	2.24	3	3.74	4.38	4.61	4.68

APPENDIX 1i TREE INCREMENT GROWTH (mm)

Tree	Circum-ference (m)	1972 2.6	16.6	28.7	20.8	6.8	21.11	28.12	4.2.73
3	3.8	–	0.5	0	–	–	–	–	0
4	2.2	2.5	2.5	2.5	2.25	0	–	–	2.0
6	4.1	0	0	0	0	0	–	–	0
8	6.4	0	0	0	1.0	0	–	0	0
10	0.17	0	0	0	2.0	2.0	–	0	4.0
14	6.0	0	0	0	0	0	0	0	0
17	5.3	0	6.0	5.5	6.5	6.5	0	0	0
19	4.55	1.0	1.0	2.0	1.0	1.5	0	5.0	6.0

APPENDIX 1 ii ROOT GROWTH

Date							Total	Av.	x/g
4.10.72	Sample wt (g)	5.61	2.36	4.75	6.32	3.10	22.10	4.42	
	No.of tips	151	69	156	176	103	685	137	31.00
	No.of galls	21	11	24	16	28	100	20	4.52
10.11.72	Sample wt (g)	3.67	6.45	7.82	11.41	8.70	31.60	6.32	
	No.of tips	80	159	187	139	200	765	153	24.21
	No.of galls	0	5	38	6	46	95	19	3.00
21.12.72	Sample wt (g)	5.56	2.17	3.41	8.94	1.44	19.35	3.87	
	No.of tips	103	54	120	192	41	510	102	76.36
	No.of galls	35	2	23	59	16	135	27	6.98
14.1.73	Sample wt (g)	1.95	4.38	7.23	5.55	3.89	23	4.60	
	No.of tips	32	121	133	147	57	490	98	21.30
	No.of galls	0	16	27	12	30	85	17	3.70
5.2.73	Sample wt (g)	6.45	6.52	6.76	7.59	1.53	29.15	5.83	
	No.of tips	207	109	174	189	31	730	146	25.04
	No.of galls	28	10	17	0	0	55	11	1.89
3.3.73	Sample wt (g)	2.58	5.67	3.69	9.41	8.40	29.75	8.95	
	No.of tips	112	173	185	348	217	1005	201	33.76
	No.of galls	12	48	10	35	55	160	32	5.38

APPENDIX 2 RATA MORTALITY - TEST FOR SIGNIFICANCE BETWEEN NORTH- AND SOUTH-FACING SLOPES

ratio alive: dead	N	S
	0.58	1.50
	1.91	0.14

$$x_1 - x_2 = 0.85$$

$$x^2 = 3.14$$

$$\overline{x} = 0.425$$

$$s = 1.9$$

$$n = 2$$

$$t = \frac{\overline{x} - u}{s/\ n} = 0.322$$

1 degree freedom

APPENDIX 3 t-TEST FOR THE SIGNIFICANCE OF SIZE DIFFERENCE IN PUPAL GALLS

Female gall diam.	Male gall diam.
350	290
385	210
435	220
300	235
390	268
352	215
371	190
345	280
360	240
345	225
400	265
342	265
316	280
363	275
354	290
378	310
380	290
380	275
318	272
364	290
7228	5235
(av. 3.61 mm)	(av. 2.59 mm)

$\overline{x} = Ex_1 - Ex_2/n = 99.65$

$s^2 = 1/19(233745-(1993)^2$

$s = 43$

$S.E. = 43/s\sqrt{n} = 9.614$

$t = \overline{x}/9.614 = 10.365$

$P < 0.001$

APPENDIX 4 A TYPICAL RECORD SHEET FOR LARVAL MEASUREMENTS

(Over 400 such measurements made. Measurements in eye-piece units at magnification shown. o = orange; y = yellow)

Gall size	Larval colour	Larval size	Head width	Sternal spatula
22	O	70x24	9	+
22	y	31x10	7	-
17	y	22x7	8	-
24	o	79x27	-	+
22	o	79x25	10	+
20	y	22x7	8	-
16	y	18x6	8	-
16	y	12x4	9	-
24	y	39x12	-	+
13	y	20x6	-	-
28	o	65x21	10	+
22	o	72x20	10	+
16	y	16x5	7	-
24	y	21x10	9	-
14	y	17x6	8	-
25	y	21x8	7	-
22	y	37x16	10	+
21	y/o	41x14	-	+
21	y	40x15	9	+
16	y	32x15	7	-
14	y	17x6	9	-
22	lo	64x17	11	+
22	lo	60x23	11	+
27	o	68x25	11	+
-	y	21x7	9	-
27	y	60x22	-	+
15	y	32x9	8	-
-	y	39x12	8	-
11	y	32x10	9	-
13	y	32x10	9	-
16	y	37x13	8	-
25	o	103x35	11	+
21	y	29x10	8	-
21	y	42x18	-	-
25	o	99x35	-	+
25	lo	62x25	-	+

APPENDIX 5. SUB-SAMPLING TRIAL

Sample	'Catch' (galls)		
	2nd instar	3rd instar	empty
1	5	2	41
2	3	3	37
3	4	3	50
4	5	7	24
5	9	16	14
6	12	11	21
Sampling total	38	41	187
Ratio (: 2nd instar)	1	1.11	4.92
Actual total	111	126	511
Actual ratio (2nd instar)	1	1.13	4.60

APPENDIX 6. ADULT EMERGENCES FROM REARING CAGES

Date	Cage					
	A		B		C	
	o	♀	o	♀	o	♀
5.2.73	1		1			
6	3	1	2			
7		1	1	1		
8	2	1	2	3	3	
9		3	1	4	1	4
12	1	1	2	8	1	3
13	4	5	6	3	2	1
15	1	4	8	5	1	1
16			1	1	1	
19		1	4	1	5	
20			1	2	1	6
21	1	2	3	2	7	4
23			1	1	2	2
27		1	2	2	1	2
28		1	1	1	1	2
1.3.73						1
2			1	1	1	1
5						2
7			1	1	1	1
8						1

APPENDIX 6i SEX ATTRACTION TRAP DATA

Trap no.	TRAPS		CONTROLS		♀/trap
	No.of males	No.of females	No.of males	No.of females	
1	40	2	6	5	1
2	33	1	3	2	1
3	13	6	4	3	6
4	31	2	8	3	3

t-test cf. males

$Ex_1 = 117$ $Ex_2 = 21$

$\overline{x} = 24$, $s = 4.4$, S.E. = 2.2, $t = 10.9$, 3 d.f.

t-test cf. females

$Ex_1 = 11$ $Ex_2 = 13$

$\overline{x} = 0.5$, $s = 11.5$, S.E. = 5.75, $t = 0.087$, 3 d.f.

t-test cf. control males & females (dispersal)

$Ex_1 = 21$ $Ex_2 = 13$

$\overline{x} = 0.2$, $s = 2$, S.E. $\doteq 1$, $t = 2$, 3 d.f., P 0.10

APPENDIX 7 TYPICAL RECORD SHEET FOR FORTNIGHTLY SAMPLES

LOCATION: SF 25.

DATE: 23.10.72.

TREE CODE: Meg.- Olymp.
1x10.
98=1cm.

[illegible]

GC = gall colour
GS = gall size
LS = larval stage

GC	GS	LS	GC	GS	LS	GC	GS	LS		NOTES
B	26 C	3	B	30CH	3.	B	35CH	3.		
B	28 CH	3	W	30	3Y	W	25	Y		MT.
W	24	Y	W	18	3Y.	B	30CH	3.		3. 8 3. 1 3. 3. 2. 1.
B	25CH	3.	B	30CH	3	B	35CH	3.		3 12 6 1 3 3. 8 4 11 4 3.
B	30CH	3	B	30CH	3.	B	27CH	3.		7. 3. 4 1
W	32	3	B	30CH	3	B	27CH	3.		6 2.
B	30CH	3	B	25CH	3.	B	27CH	3.		5. 3 3. 16. 8 10 8
W	21	Y	B	23CH	3.	B	26CH	3		7. 3. 4 –.
B	31CH	3	B	29CH	3.	W	31	3Y		MORT
W	38	Y	B	29CH	3.	W	24	Y		21. 33. 20 2 21 30 34 21 27 25.
B	30CH	3	W	18	Y	W	30	Y		23. 31 33 3 20. 31 23 22 25. 22
W	35	3Y	W	22	Y	B	30CH	3		18 26. 22.
B	34CH	3	B	25CH	3	W	36	3Y.		1 8cm.
W	22	Y	W	31	Y	B	33CH	3.		1 8cm 1 "
W	35	3Y	B	25CH	3	B	23CH	3		1 " 1 " 1 "
B	32CH	3	B	26CH	3.	W	36	3Y.		1 " 1 "
B	32CH	3	W	31	3Y	W	32	3Y		1 " 1 " 1 " 1 "
B	32C.	3	W	31	3Y.	W	30	3		2 pupal ski
B	23CH	3.	B	25CH	3.	B	30CH	3.		
B	31 CH	3	W	25	Y.	B	22CH	3		
B	36CH	3	B	31CH	3.	B	32CH	3.		
W	31	3Y	B	29CH	3.	B	28CH	3Y.		cells 2. with
B	25CH	3.	B	30CH	3.	LB	23	3Y		no sign of c
B	35CH	3	B	28CH	3	B	34CH	3.		
B	33CH	3.	B	27CH	3.	W	26	Y		
B	34CH	3	B	23CH	3.	W	30	3Y		

APPENDIX 8 ANALYSIS OF VARIANCE - HORIZONTAL DISPERSION

No.of galls (25 samples/tree)

Tree	-	-	-
	359	97	60
	14	62	223
	65	140	5
	60	43	16
	2	346	10
	13	78	43
	650		1
	115		14
	70		20
	3		4
	8		
Total	1365	766	396
$E(x^2)$	577 809	160 502	6 272
$E(x)^2$	1 863 225	586 756	56 332
x	54.6	30.64	15.84
S^2	20 970	5 709.65	2 085.8

	Sum squares	Df	Mean square	Variance ratio
Trees	19 128.88	2	9564.44	-1.0051
Residual	-685 171	72	9516.26	
Total	704 300			

APPENDIX 8i DATA FOR INTER-TREE DISPERSION

Tree no.	Sample fresh wt	Dry wt	% soil moisture	Aspect	Circumference (m)	Altitude (ft)	Infestation	Health
1	229.2	165.4	63.8	S	6.1	1200	9	++
2	237.4	173.8	63.6	S	4.1	1200	5	++
3	204.5	152.5	52.0	S	3.8	1260	3	++
4	231.3	184.9	46.4	N	2.2	1300	0	++
5	231.3	184.9	46.4	N	0.5	1290	0	++
6	238.7	183.7	55.0	0	4.1	1280	8	++
8	190.2	146.3	43.9	S	6.4	1400	7	++
9	165.6	124.6	41.0	S	5.6	1400	5	++
10	172.2	150.5	21.7	0	0.17	1440	1	++
11	163.1	138.2	24.9	N	3.4	1430	3	++
12	182.2	152.0	30.2	N	4.5	1400	4	++
14	194.6	147.0	47.6	S	6.0	1500	7	+
17	201.5	157.2	44.3	0	5.3	1500	3	+
16	163.1	130.8	32.3	S	3.8	1490	3	+
15	156.4	132.9	23.5	S	4.7	1490	3	+
19	167.3	136.8	30.5	0	4.55	1550	6	+
18	221.3	176.0	45.3	N	4.4	1480	6	++
21	251.5	244.6	7.1	N	4.0	900	0	++
22	132.2	125.0	7.2	N	2.5	1000	0	+++
*30	250.0	219.8	30.2	N	1.45	935	0	+++

* terrestial tree in open field adjacent to the study area

infestation ranked 0-10

tree health ranked + - +++

aspect: S = south-facing N = north-facing 0 = west-facing

circumference at breast height

APPENDIX 8ii DISPERSION DATA

(weights in g)

TREE 1

Sample co-ords	Fresh wt	Dry wt	Ash wt	% soil moisture	% soil org. content	Metrosideros root wt	M.robusta root wt	No.of galls	Galls/g root	Glass wt
03	17.5934	13.6973	12.6905	52.2	28.2	6.4	>3	359	6	10.1311
09	19.3593	14.9557	14.0424	52.0	22.4	0.05	<1	0	0	10.8844
15	18.3108	13.8966	13.0921	56.2	23.4	0.1	<1	0	0	10.4531
34	10.6478	8.4324	7.6996	57.0	43.8	3.6	>3	14	3.9	6.7629
32	19.4847	15.1515	14.0415	49.0	24.6	3.0	>3	65	21.7	10.5427
46	12.5907	9.6900	9.1043	49.8	19.8	0.1	<1	0	0	6.6443
43	14.1643	10.6570	9.7425	47.2	23.3	0.76	<1	6	7.9	6.7616
33	19.5120	14.8695	13.6584	50.3	26.1	1.1	<2	60	54.4	10.2795
61	11.8822	9.4300	8.7465	47.6	25.3	2.2	<3	2	0.91	6.7334
31	19.7335	14.3407	14.1476	49.3	26.4	2.2	>3	13	59	10.8235
42	15.2542	12.1470	10.6736	62.5	78.9	12.5	>5	650	52	10.2795
71	10.8458	15.6418	14.3634	46.6	26.6	0.8	<1	0	0	10.8247
70	19.6846	15.5370	14.4347	46.7	23.3	0	0	0	0	10.7985
53	13.1324	10.1848	9.5005	45.9	19.7	0.3	<1	0	0	6.7079
72	14.0200	10.7074	9.8708	45.4	21.0	0.15	<1	0	0	6.7230
78	12.7847	9.6395	9.0140	52.1	21.3	0.25	<1	0	0	6.7340
98	18.4918	14.3580	13.4898	49.9	21.0	0	0	0	0	10.2145
63	19.1627	14.0353	13.9881	49.9	24.4	0.1	<1	0	0	10.7499
91	12.5480	9.4941	8.8326	52.0	23.5	2.0	<2	115	57.5	6.6800
54	13.6685	10.2104	9.4255	49.5	22.3	0	0	0	0	6.6883
90	18.7894	14.5100	13.3593	51.6	28.6	1.7	<2	70	41	10.4923
93	13.3627	10.1524	9.4670	49.0	20.5	0.5	<1	3	6	6.8139
79	10.7478	8.6235	8.1680	53.0	24.4	0	0	0	0	6.7424
05	18.6049	14.4573	11.3232	52.0	19.5	0.5	<1	8	16	10.6352
37	18.2500	14.0939	13.2252	53.9	24.5	0	0	0		10.5427

APPENDIX 8 (iii)

TREE 6

Sample co-ords	Fresh wt	Dry wt	Ash wt	% soil moisture	% soil org. content	Metrosideros root wt	M.robusta root wt	No. of galls	Galls/g root	Glass wt
42	17.1744	14.0709	13.0743	48.2	29.3	0	0	0	0	10.7366
62	19.7991	16.5324	15.8087	33.9	11.4	0	0	0	0	10.1666
32	17.3892	13.8295	12.8834	52.7	29.6	0	0	0	0	10.6370
16	21.0090	17.3311	16.5159	36.0	12.5	0.4	<1	0	0	10.8105
46	14.3540	12.0869	10.9151	58.9	74.1	0	0	0	0	10.5010
00	12.8144	11.8426	10.8942	48.2	91.0	1.6	<2	0	0	10.8000
83	18.9825	15.3406	14.4175	43.2	19.2	1.8	<2	0	0	10.5438
33	19.5602	15.7944	14.7696	41.3	19.1	0.6	<1	0	0	10.4408
31	17.7023	14.4209	13.5470	46.0	33.0	0.5	<1	0	0	10.5711
08	16.1910	12.8241	11.0008	58.1	34.4	1.0	<2	97	97	10.4006
11	19.4730	15.0896	15.8241	42.1	14.7	0	0	0	0	10.8021
19	19.8290	16.0845	15.4085	40.7	12.6	0	0	0	0	10.7095
56	20.7594	17.0966	16.1983	36.4	14.0	0	0	0	0	10.6893
05	18.3697	15.2034	14.3744	40.6	17.9	5.2	>3	62	11.6	10.5759
04	14.0297	11.7557	10.7648	60.5	66.7	7.5	>3	140	18.6	10.2703
34	18.1275	14.6747	13.8294	46.1	21.6	0.4	<1	0	0	10.7690
92	20.2426	16.7797	16.0886	36.0	11.2	0.01	<0	0	0	10.6111
76	21.5560	18.8310	18.1386	30.5	8.3	0	0	0	0	10.5000
24	15.8510	12.6692	11.7434	59.4	43.6	1.9	<2	43	22.5	10.4866
45	16.0526	13.3266	12.2965	48.7	35.9	0	0	0	0	10.4565
38	17.7413	14.1762	13.1831	52.6	30.9	0.2	<1	0	0	10.9627
07	20.3929	16.9831	16.4291	34.1	8.4	0	0	0	0	10.3890
48	15.6200	12.2999	11.3363	66.2	56.8	5.5	>3	346	65.4	10.6049
14	17.3843	14.1057	13.3740	45.5	18.6	1.9	<2	78	43.4	10.1761
86	21.1197	17.8722	17.2575	30.5	8.3	0	0	0	0	10.4756

APPENDIX 8 (iM)

TREE 8

Sample co-ords	Fresh wt	Dry wt	Ash wt	% soil moisture	% soil org. content	Metrosideros root wt	M. robusta root wt	No. of galls	Galls/g root	Glass wt
44	14.1577	11.9577	11.0024	57.7	59.3	0.8	<1	60	75	10.3454
62	15.5534	12.3921	11.1980	32.2	65.7	0.3	<1	0	0	10.6416
67	18.9515	15.4144	14.5948	42.5	17.1	0	0	0	0	10.6223
37	14.3286	11.6593	10.7913	73.5	79.3	14.0	>3	223	16	10.6969
23	18.3035	14.5099	13.5732	50.6	25.3	0	0	0	0	10.8017
16	21.7477	17.2920	16.4210	40.9	13.5	0	0	0	0	10.8529
07	20.0415	15.7109	14.8673	45.6	16.4	0	0	0	0	10.5535
52	15.0000	12.2100	11.1836	68.9	81.3	1.2	<2	0	0	10.9481
47	21.4583	17.1829	16.2372	39.7	14.8	0	0	0	0	10.7754
38	19.3432	15.3695	14.4759	38.2	18.0	0.2	<1	5	25	10.3985
10	14.6482	11.5380	10.5823	73.3	84.2	1.3	<2	16	12.3	10.4023
04	22.1158	17.9739	17.1773	36.0	10.8	2.3	<3	10	4.35	10.6262
50	14.7115	12.4761	11.1979	57.8	78.4	4.4	>3	43	9.8	10.8448
17	21.0100	16.5166	15.6809	64.2	14.7	0	0	0	0	10.8366
43	13.8689	11.9960	10.8757	55.6	74.7	0.7	<1	1	1.4	10.4994
-	19.9227	15.4752	14.4302	46.7	20.6	0	0	0	0	10.4089
46	14.4024	11.4465	10.6633	38.6	16.6	0	0	0	0	6.7349
09	14.2255	10.7675	10.1551	45.6	14.8	0.2	<1	14	70	6.6378
-	10.3337	7.7847	6.9382	57.0	44.1	0	0	0	0	5.8632
-	32.7182	24.7319	23.5076	51.2	16.1	0	0	0	0	17.1294
74	25.2914	20.4800	19.1490	62.8	46.7	0.4	<1	20	50	17.6284
93	30.9100	24.5876	23.1371	42.9	14.5	0	0	0	0	15.7261
-	31.3207	24.5830	23.2655	44.5	15.8	0	0	0	0	16.1987
71	27.3178	21.7100	20.4509	50.3	22.7	0.8	<1	4	5	16.1593
83	37.8558	30.3513	29.0072	40.0	11.6	0	0	0	0	19.1044

Total root weight from Trees 1,6,8 = 93.42 g

APPENDIX 8 v VERTICAL DISPERSION - SUMMARY OF DATA

Gall no.	Root wt (g)	Depth (cm)	Org.matter %	Gall no.	Root wt (g)	Depth (cm)	Org.matter %
62	0.5	2	65	72	0.94	6	92
5	0.12	4	51	2	0.05	8	64
0	0.1	6	52	3	0.05	2	64
0	0.24	8	52	1	0.05	4	56
93	0.9	2	92	0	0	6	56
29	0.42	4	94	0	0	8	56
13	0.25	6	70	5	0.04	2	77
0	0.1	8	55	0	0	4	60
0	0	10	51	0	0	6	52
350	2.4	2	99	0	0	8	49
71	1.0	4	98	24	0.25	2	76
25	0.5	6	89	23	0.32	4	51
3	0.1	8	77	0	0	6	51
0	0.1	10	66	0	0	8	50
111	1.0	2	98	187	2.4	2	93
55	0.45	4	97	98	1.0	4	92
00	0.2	6	68	15	0.2	6	70
0	0	8	56	0	0	8	68
0	0	10	55	0	0	10	55
49	0.5	2	68	0	0.4	2	73
0	0.1	4	49	0	0	4	65
0	0.1	6	48	0	0	6	51
0	0	8	48	0	0	8	49
0	0	10	48	0	0	10	50
256	3.1	2	97	43	0.6	2	64
109	1.4	4	92	20	0.3	4	63
32	0.6	6	87	0	0.1	6	61
0	0.5	8	71	1	0.1	8	53
0	0	10	67	0	0	10	55
39	0.8	2	47	220	2.0	2	96
6	0.44	4	45	120	0.9	4	94
0	0	6	49	64	0.6	6	90
0	0	8	47	0	0.3	8	71
602	4.5	2	94	0	0	10	65
189	2.25	4	93				

APPENDIX 9 SUCTION TRAP DATA

Date	Start (hr)	Stop (hr)	Duration (min.)	Wind speed (m/min.)	No.of cecida	No.of *P.radicis*
23.2.72	1745	1946	115	0.6	17	–
24.2	1812	1842	30	100	0	
25.2	1725		45	1.3	4	
27.2	1730	1921	110	0.6–59	33	
28.2	1822	1904	100	0.6	35	
29.2	2038	2141	63	0	9	
"	2141	2249	65	2	9	
"	2252	0000	68	0	8	
1.3.72	0955	1055	60	4	10	
"	1132	1202	30	1.3	2	
"	1230	1301	31		8	
"	1330	1430	60		7	
"	1500	1530	30		7	
"	1600	1700	60		1	
"	1701	1801	60		5	

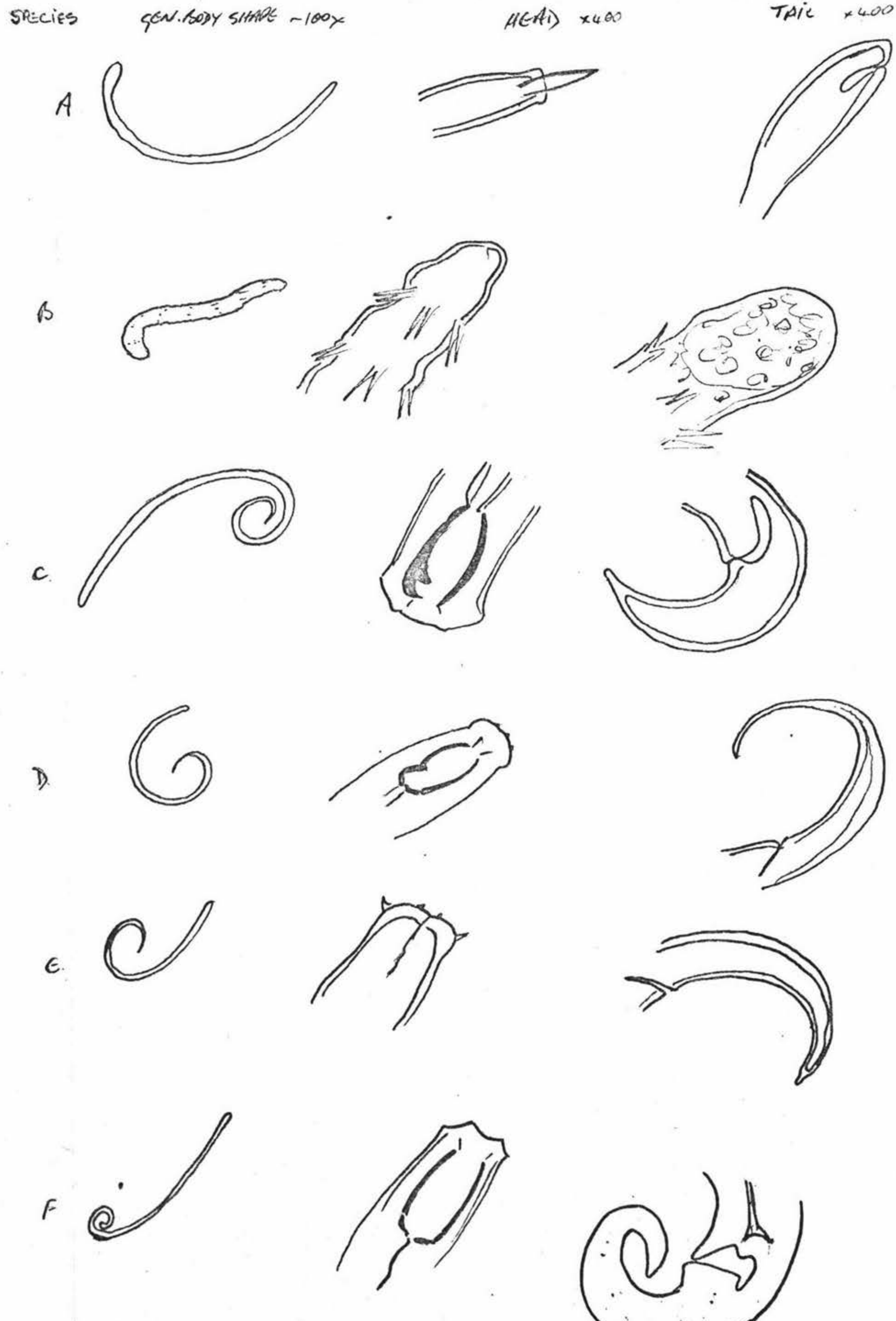
MALAISE TRAP DATA (1972)

Date	No.of cecids	No.of *P.radicis*
6.1.72		
21.1	37	0
30.1	22	0
4.2.72	10	0
11.2	6	0
27.2	6	0
3.3.72	4	0

APPENDIX 10 DESICCATION DATA

	WEIGHT (g) OF				
	larvae + glass	gall + glass		root + glass	root
Initial weight	6.62820	24.2540	23.9763	38.0318	2.0041
Weight after (min.) 5	6.62816	24.2305	23.9622	37.9830	1.9549
10	6.62813	24.2078	23.9392	37.9363	1.9069
15	6.62812	24.1859	23.9172	37.8940	1.8638
20	6.62812	24.1658	23.8969	37.8520	1.8208
25	6.62812	24.1458	23.8766	37.8100	1.7777
30	6.62812	24.1258	23.8563	37.8670	1.7343
35	-	24.1058			
280	-	23.2776			
1040	-	20.9336		36.6938	
Dry weight	6.62800	19.4905	19.1727	36.4734	-
Glass weight	6.62796	18.4743	18.1749	36.0522	-
Fresh wt tissue	0.00024	5.7797	5.8014	1.9796	2.0041
Dry weight tissue	0.00004	0.9962	0.9978	0.4212	0.4222

APPENDIX 11 KEY FOR THE IDENTIFICATION OF NEMATODES




G
400x
H
I
J (or H?)
K. small
no teeth
L. small
no teeth
M

APPENDIX 12 SPECIES ASSOCIATION DATA - THE ANIMAL COMMUNITY ABOUT M. robusta

Sample	1st instar larvae	galls	Nematode species	Mite species	Other arthropods
TREE 17					
A	-	-	GO	R	ma Ba Ae Rb
B	+	+	DGCAE	GCOBA1	Ca Ma Ha Dabe Ae Pa Xa
C	-	+	JMC	FG	Cab Ma Da Pa Gbc Bag Rb
D	-	-	CAFP	FHIJKG	Ca Bagh Mb Pa Wa Gc Ra
E	-	+	JFC	FKL	Ca Mab Bag Dcg Ae Pa Sb Xab
TREE 7					
A	+	+	JKC	FGIJM	Ma Rc Ha Dc Aae Pa Beg Xa Gbe
B	+	+	MCJ	EIGNOPQ	Mab Rb Ha Bbf De Sa Xa G
C	+	+	ACE	ISPZC	Cab Mab Rac Dbe Fa Pa La Sb Gab
D	+	+	ACE	FSQABMT	Cab Ma Ra Dabcf Pa Xa Ga
E	-	-	BNEAG	ACDVD1	Ca Ub Rb Dc Mb Pa Eab Lab Sb Xa
TREE 1					
A	-	-	BGEAN	ASHEYTGI	Ca Rabc Dcd Fb Bcd Eb Sbc
B	+	+	ABCDEF	IM	Ma Dbcf Aed Bg Eab Xa
C	-	+	AEDQF	ASEOF	Ca Ub Rc Ha Dac Ac Be Sbe Xa Wa Ia
D	+	+	EACH	AINKD1	-
E	+	+	BMAECD	EWX	Ca Mab Rb Hc Ba Ab Pa Eab Sa

Sample	1st instar larvae	Galls	Nematode	Mite species	Other arthropods
TREE 6					
A	+	+	FL	RFMIDO	Ma Rb Dcf Aed Pa La Sb Xa Gb
B	-	+	CHJGKL	-	Ub Rb Ha Dc Fc Aed La Sb Xa
C	-	+	FGC	-	Ca Rc Dce Ad Pa Bf Wa
D	-	+	ACBD	AESQ	Ca Ra Dc Mb La
E	+	+	CHAMN	-	-
TREE 8					
A	-	-	DMOE	-	Ub Sc Dac Mb
B	-	+	EAIFD	-	Ma Dac Fa Ea W
C	-	-	JOMFDBE	FACUJ	Ca Mab Ub Aa Pa
D	+	+	KAGEF	ABCDE	Ca Ma Uabc Ra Ha
E	-	+	CBAMD	-	Ca

Mites

Cryptostigmata (omnivorous) B C D E F G H L P R V Y Z Di
Trombidiiformes (predators) A I M N Q S T W X
Parasitiformes (carnivorous-ditritophagous) O U
Metastigmata (ticks) K
(Nomenclature of Evans, G.O., 1955)

Other Arthropods

C Crustacea - amphipods sp. a & b
M Myriapoda - a = centipede b = millipede
B Coleoptera - a & b = larvae c-h = adults
R " adult Staphilinidae rove beetles
U " " Curculionidae weevils
E " larval Elateridae click beetles
H Hymenoptera - a = ceraphronid parasite of <u>P.radicis</u>
A " ants
D Diptera - larvae
F " adults
G Hemiptera - a = pseudococcid b = assassin c = margaroidea
X Apterygota - a = symphylan b = dipluran
P Chelonethi - pseudoscorpion
S Arachnida - Toxopidae groundhunting spiders
W Annelida - earthworms
I Mollusca - snail

APPENDIX 13 DATA FOR THE CERAPHRONID PARASITISM OF P.radicis

Sample	No.of galls	2nd instar	3rd instar		Pupae		No.of parasites
			whole	fungi	whole	exuviae	
a71	5	-	-	-		1	
a34	14	-	1	-		1	2
a32	65	3	12	6	1	9	2
a61	2	1	-	-		1	
a43	6	1	-	-	1		
a05	8	1	1	-		1	
a31	13	-	-	1		5	
a93	3	-	-	-			
a33	60	2	10	-	3	7	3
a92	70	2	-	-	2	1	3
c74	20	2	-	-			1
c10	16	-	-	-			
b08	97	6	1	2	1	4	2
a03	259	19	27	5	8	12	9
b05	62	9	3	1		1	
b04	140	19	12	4	1	9	3
c50	43	1	1	1			
c37	173	33	23	6	4	8	1
c09	14	-	1	-			
c38	5	-	-	-		1	
c04	10	2	1	1			
c62	-	-	-	-			
c71	4	2	-	-			
b24	43	7	3	2	2		
c44	60	2	-	1		1	
a91	115	1	2	-			
b14	78	7	1	1	3	4	1
c43	1	-	-	-			
b48	296	38	23	2	6	15	5
a42	550	47	61	13	8	26	9
Total	2232	205	183	46	46	107	41

% parasitism = 21.8% (pupae), % fungus on 3rd instars = 20%

APPENDIX 14 LIFE TABLE

From sample

188 pupae/83.42 g root

% pupal mortality (parasitism + other) = 22.34%

∴ no. of live pupae/g root $= \frac{146}{93.42} = 1.56$

Total maximum fecundity (eggs/g root) $= \frac{1.56}{2} \times 116 = 90.47$*

eggs/11.05 g = 1000

% 2nd instar mortality = 11.29%

% 3rd " " = 6.79%

<u>given</u> % mortality in a stage = a

and no. entering following stage = m

$$\text{let } b = x - y \qquad \ldots (1)$$

$$\text{then } a = \frac{y}{x} \times \frac{100}{1} \qquad \ldots (2)$$

$$ax = 100y$$

$$x = \frac{100y}{a}$$

substitute for x in . . . (1)

$$b = \frac{100y}{a} - y$$

given a & b ∴ solve for y

substitute y in equation . . . (1) to find x.

By this method the numbers entering the 3rd instar and 2nd instar can be found concurrently.

% mortality of eggs and 1st instar from 1000-no. entering 2nd instar expressed as a percentage of 1000.

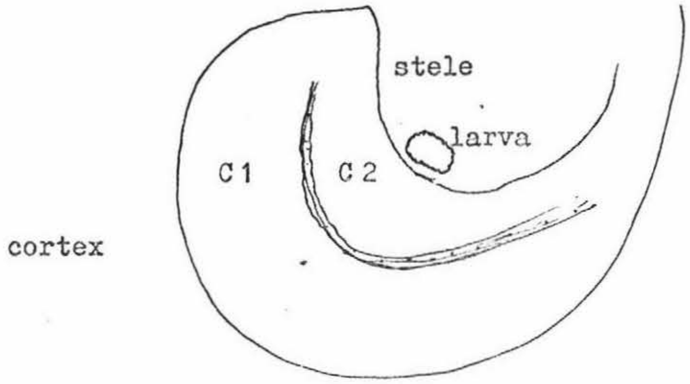
---

* assuming a sex ration of 1

APPENDIX 15 HOST SPECIFICITY - MEAN LEAF DIMENSIONS (mm)

Locality	Leaf length	Leaf breadth	Galls
study area (S.W. Ruahines)	50.7	17	-
	47.0	16	-
	30.0	14.3	+
	30.0	11.3	+
	29.7	10.3	+
	39.5	12.5	+
	49.7	15.5	+
	30.5	11.5	+
	50.0	15.2	+
	31.7	13.2	+
	41.2	15.0	+
	36.7	15.0	+
	35.5	15.0	+
	35.5	11.0	+
	28.5	12.8	+
	45.0	14.0	+
	35.5	13.0	+
Makoia Is.	42.9	24.5	-
	101.4	25.1	-
	48.1	17.1	-
	55.2	18.6	-
	73.0	29.1	-
	83.9	33.2	-
Pohutukawa Bluff	65.0	19.5	-
	48.0	16	-
	47.0	20.5	-
	52.8	17	-
	59.0	24	-
	71.0	26	-
	63.0	22.5	-
	65.5	17	-
	53.0	19	-
	43.5	18	-
	62.0	18	-
	54.0	21	-
	43.0	13	-
	76.0	23	-
	49.0	19	-
	53.5	17	-
	41.0	16	-
Wanganui	68.2	23	-

APPENDIX 16 GALL STRUCTURE - CORTICAL CELL DIMENSIONS (e.p.u.)



	C1					C2				
(epidermis)	(length x breadth)					(length x breadth)				
	25x9	15x7	19x9	21x8	21x4	-x2	10x2	11x4	20x2	14x4
	24x7	12x8	27x6	18x11	19x9	-x2	13x4	23x5	14x4	19x2
	29x7	16x12	15x8	25x8	23x7	-x2	-	22x3	22x4	18x2
	23x14	11x11	18x7	23x7	26x6	24x4	-	30x4	22x4	23x2
	35x8	19x10	17x9	24x8	24x10	42x2	11x5	30x5	40x4	37x7
	31x7	11x18	17x10	30x11	30x10	31x5	16x4	30x6	40x6	12x7
cortex	30x13	14x10	23x9	25x5	22x10	22x5	-	20x7	17x3	16x4
	21x16	30x10	30x11	23x7	19x8	28x5	21x4	33x5	24x6	21x6
	17x12	21x12	20x10	30x15	20x12	26x6	34x6	32x9	17x9	22x9
	15x20	14x14	15x15	25x10	34x9	28x6	28x7	20x6	16x8	21x10
	35x10	28x10	25x15	15x11	25x15	23x7	35x9	14x5	25x5	30x6
	12x6	14x11		32x6	21x14		24x13	20x4	30x5	22x6
	25x13	22x8		26x8	21x13		18x8		25x10	30x7
		12x8		30x11						
(stele)		13x5								

Normal root	(length x breadth)					
(epidermis)	22x6	24x13	21x8	14x6	10x6	25x5
	12x5	25x5	24x8	20x12	24x6	25x10
cortex	25x18	12x18	15x18	25x8	24x9	24x7
	10x15	21x9	20x12	22x17	25x16	15x11
	9x5	24x9	22x6	16x8	21x7	15x10
(stele)	5x7	17x7	11x8	20x5	20x7	18x7

APPENDIX 17 A TYPICAL RECORD SHEET FOR THE RESPIRATION EXPERIMENTS
(20°C)

Tissue	Fresh wt	Initial	Time (min.)					
	(g)	(ul)	10	20	30	40	50	60
blank		104.7	104.7	104.7	104.7	104.7	104.7	107.2
White galls	3.88953	100.2	148.5	196.1	244.0	293.4	336.8	384.4
"	4.48444	105.1	156.1	207.9	260.5	312.1	362.2	413.0
roots	1.39570	120.3	138.8	155.9	176.0	193.1	210.7	225.5
blank		102.3	103.2	105.2	106.3	106.3	107.9	107.9
brown galls	2.12667	110.9	113.1	114.7	115.3	115.3	116.1	116.1
roots	1.09540	100	114.1	132.7	146.3	160.7	176.8	189.3
brown galls	2.51304	104.8	121.5	139.2	155.5	170.2	187.2	201.2
white galls	3.19490	101.8	114.9	131.7	146.8	162.3	176.8	191.9
blank		121.8	121.8	124.3	126.2	127.7	129.0	129.0
white galls	2.87156	113.8	147.4	183.5	218.9	252.3	285.7	320.8
blank		120.1	120.1	121.3	122.0	122.0	122.0	123.3
		130.0	205.1	205.1	241.6	278.1	315.6	350.3
white galls	3.56683	12.42	167.5	209.3	251.8	294.5	334.0	374.5
roots	1.97955	114.8	140.4	165.0	189.0	212.5	233.0	255.7
white galls	2.68845	130.0	154.4	178.2	201.7	224.9	247.8	269.9
roots	0.97456	139.5	134.8	205.0	244.4	299.7	338.5	386.5
roots	1.19389	133.0	155.8	180.9	201.4	223.5	224.7	265.2
brown galls	2.79803	116.5	146.1	169.3	189.0	209.5	228.5	248.1
blank		136.4	136.4	137.8	137.6	137.6	139.6	140.4

(Average of blank recordings is subtracted from each tissue recording at each time interval.)

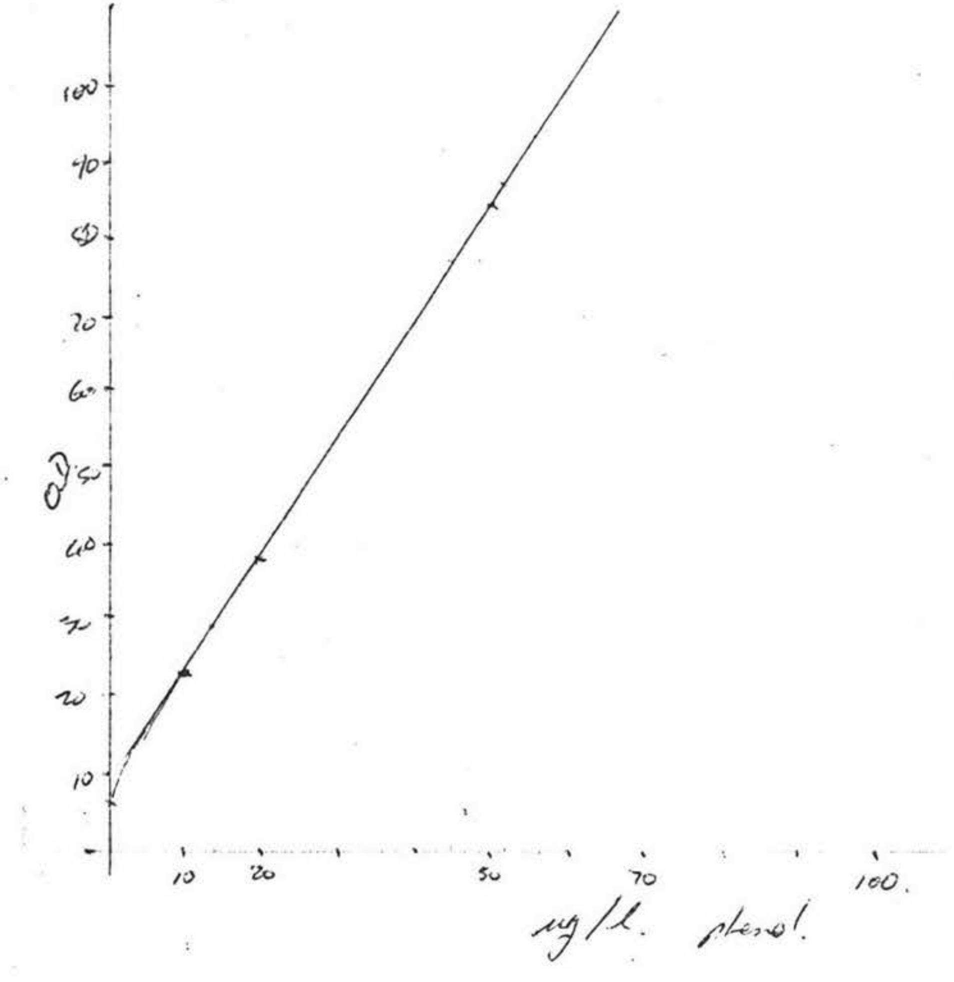
APPENDIX 18 GALL CHEMISTRY

Tissue	Fresh wt. g	Dilution	Phenol equiv. µg/l	Dilution	O.D. at 550 mµ	Dilution	Tannin equiv. mg/ml
ROOT	0.7315	20	40	10	14.2	10	0.6
	0.49	20	18	5	11.4	10	0.3
	1.045	20	24	10	37	10	0.7
GALL	1.5926	50	40.5	100	13.5	50	0.6
	0.818	50	26	100	6	50	0.3
	1.416	50	29	100	15	50	0.4

' STANDARD CURVE ' FOR TANNIN

Concentration mg/ml	Precipitation of haemoglobin
0.3	0 none
0.5	++
0.6	+++
0.8	+++++ complete

18 i STANDARD CURVE FOR PHENOL



Standard curve. phenol.
10 - 70 µg/l. OK.

1ml soln.
0·5ml Folin-Ciocalteau. (dil 1:1 H_2O)
wait 3min
2·0ml 20% Na_2CO_3
2·5ml H_2O.

if H_2O dilution increased
to 10ml prob be OK
for 10 - 100 µg/l.

1 hr avail

read at ~~725 mµ~~
760 mµ

ALLAN, H.H. 1961: Flora of New Zealand I. NZ Government Printer, Wellington. 1085 pp.

ALSTON, R.E. & HEMPEL, K. 1964: Chemical documentation of interspecific hybridisation. J. Heredity 55: 267-9.

ANDERSON, J.A.J. 1935: Immature and adult stages of *Oligotrophus oleareae*. Proc. Zool. Soc. 1935.

——— Morphology of a new *Dryomgia* sp. Proc. Zool. Soc. 1935.

ASTON, B.C. 1918: The indigenous tans and vegetable dye stuffs of New Zealand. NZ I. Ag. 17: 136-40.

BALASABROMANIAN, M. & RANGASWAMI, G. 1962: The presence of indole compounds in nematode galls. Nature (Lond.) 194: 774-5.

BALGOOY, M.M.J. van. 1966: The Myrtaceae in Pacific Plant Areas. v. 2. Blumea Suppl. 5.

BARNES, H.T. 1937: Check list of New Zealand Cecids. Trans. roy. Soc. NZ 67: 115-21.

——— 1946-56: Gall Midges of Economic Importance. v. 1-7. Crosby, London.

BATE-SMITH, E.C. 1973: Haemanalysis of tannins: the concept of relati astringency. Phytochem. 12: 907.

BEDDIE, A.D. 1953: Root behaviour in *Metrosideros*. Wellington Bot. Soc. Bull. 26: 2-6.

BERLAND, L. 1935: Premiers resultants de mes recherches en avion sur la fauna et aa flora atmospheriques. Ann. Soc. Ent. Fra. 104: 73-96.

BERNAYS, E.A. & CHAPMAN, R.F. 1970: Experiments to determine the basis of food selection by *Chorthippus parallelus* in the field. J. Anim. Ecol. 39: 761-76.

BETTS, M.W. 1919: Notes on the autecology of certain plants of the peridotote belt, Nelson. I - Structure of some plants. Trans. Proc. NZ Tech. Inst. 51: 136-56.

BIRD, A.F. 1962: The inducement of giant cells by *Meliodygne juvenica*. Nematol. 8: 1-10.

BORROR, D.J. & De LONG, D.M. 1964: An Introduction to the Study of Insects. Holt Reinhart, NY.

BROOKER, S.G., CAIN, E.F. & CAMBIE, R.C. 1963: A New Zealand phytochemical register. Pt 1. Trans. roy. Soc. NZ, Gen. 1: 61-87.

BROWER, L.P. 1969: Ecological chemistry. Sci. Am. 220(2): 22-9.

BROWN, S.A. 1961: The chemistry of lignification. Science 134: 305.

BURSTROM, H.G. & SVENSSON, S.B. 1972: Hormonal regulation of root growth and development, in Proc. Adv. Study Inst. izmer (Kaldeway & Larder eds.), Verlag Chemie, Weinheim.

CARTWRIGHT, W.B. 1922: Sexual attraction of the female Hessian fly. Phytophaga destructor. Can. Ent. 54: 154.

CHALLICE, J.S. 1972: The phenolics of Pyrus interspecific hybrids. Phytochem. 11: 3015-18.

CHILVERS, G.A. 1972: Tree root pattern in mixed eucalypt forest. Aust. J. Bot. 20: 229-34.

CHITTY, D. 1957: Self-regulation of numbers through changes in viability. Cold Spring Harb. Symp. quart. Biol. 22: 277-80.

CLARK, L.R. et al. 1967: The Ecology of Insect Populations. Methuen, Lond. 232 pp.

CLAUSEN, C.P. 1962: Entomophagous Insects. McGraw-Hill, NY. 688 pp.

CLOWES, F.A.L. 1969: Hormones and plant development, in Proc. Adv. Study Inst. izmer (Kaldeway & Larder eds.).

COCKAYNE, L. 1927: New Zealand Plants and their Story. Government Printer, Wellington.

COOPER, R.C. 1954,1958: Pohutukawa x rata: variation in Metrosideros on Rangitoto Is., New Zealand. Rec. Auck. Inst. Mus. 4(4): 205- 5(1,2): 13-40.

COUPER, R.A. 1953: Metrosideros pollen from Eocene. NZ Geol. Surv. Paleo. Bull. 22. 77 pp.

CRUICKSHANK, I.A.M. & PERRIN, D.R. 1964: Pathological function of phenolic compounds in plants, in Biochemistry of Phenolic Compounds (Harborne, G.B., 1964).

DAUBENMIRE, R.F. 1968: Plant Communities - A Textbook of Plant Synecology. Harper & Row, NY

DAWSON, J.W. 1968: An analysis of flowers and fruits in New Zealand Metrosideros. NZ J. Bot. 6: 43-55.

——— 1968: Vegetative buds of New Zealand species of Metrosideros. NZ J. Bot. 6: 240-2.

——— 1970: Pacific capsular Myrtaceae. The Metrosideros complex: M.collina group. Blumea 18: 441-5.

DETHIER, V.G. 1954: Evolution of feeding preferences in phytophagous insects. Evol. 8: 33-54.

——— 1970: Chemical interactions between plants and insects, in Chemical Ecology (Sondheimer & Simone eds.), Ac. Press, NY.

DIELMAN, F.L. 1969: The effects of gall midge infestation on plant growth and growth regulating substances. Ent. Exp. Appl. 12: 745-9.

DIXON, A.F.G. 1971: Stabilisation of aphid populations by an aphid induced plant factor. Nature 227: 1368-9.

DUMBLETON, L.G. 1967: Winter dormancy in New Zealand biota and its paleological implications. NZ J. Bot. 5: 211-22.

EDWARDS & PRITCHARD. 1968: New classification of former S.F. Heteropezinae. Ann. Ent. Soc. Am. 53: 305-16.

ELDER, N.L. 1965: Vegetation of the Ruahine Ranges. NZFS Tech. Pap. 45.

ELLIS, CARLISLE, D.B. & OSBORNE, D.J. 1965: Desert locusts: sexual maturation delayed by feeding on senescent foliage. Science 149: 546-7.

ENGELBRECHT, L. 1971. Cytokinin activity in larval infected leaves. Biochem. Physiol. Pflanzen 162: 9-27.

ENGELBRECHT, L., ORBAN, U. & HEESE, W. 1969: Leaf-miner caterpillars and cytokinins in the green islands on autumn leaves. Nature 223: 319-21.

EVANS, G.C. 1972: Quantitative Analysis of Plant Growth. Blackwell, Oxford. 734 pp.

EVANS, G.O. 1955: Identification of terrestial mites, in Soil Zoology (McKeven, D.K., ed.). Butterworth, Lond.

FARKAS, G.L. & KIRALEY, Z. 1962: The role of phenolic compounds in the physiology of plant diseases and disease resistance. Phytopath. 44: 105-55.

FELT, E.P. 1925: Key to gall midges. NY St. Mus. Bull. 257: 1-239.

——— 1936: Relation of insects and plants in gall production. Am. Ent. Soc. Am. 29: 694-700.

——— 1940: Plant Galls and Gall Makers. Comstock Pub., Ithaca, NY. pp. 1-364.

FENNY, P.P. 1968. The effects of oak leaf tannins on larval growth of the winter moth Oparophtera brumata. I. Insect Physiol. 14: 805-17.

——— 1970: Seasonal changes in oak leaf tannins and nutrients as a cause of spring feeding by winter moth. Ecol. 51: 565-81.

FENNY, P.P. & BOSTOCK. 1968: Seasonal variation in tannin content. Phytochem. 7: 871-80.

FRAENKEL, G. 1953: The nutritional value of green plants for insects. Trans. 9th Int. Cong. Ent., Amsterdam 1951, 2: 70-100.

——— 1959: The raison d'etre of secondary plant substances. Science 129: 1466-70.

——— 1969: The evaluation of our thoughts on secondary plant substances. Ent. Exp. Appl. 12: 473-86.

FRANKLIN, D.A. 1967: Synecology of Tararua indigenous forests. NZFS Tech. Pap. 53.

GIESE, R.L. & BENJAMIN, D.M. 1959: The biology and ecology of the Balsam Gall Midge in Wisconsin (Dasyneura balsamicola). For. Sci. U.S.

GILMOUR, J.W. 1966: The pathology of forest trees in New Zealand. NZFS, FRI

GLOCK, W.S. & AGERTER, S. 1963: Anomalous patterns in tree rings. Endeavour 9: 23.

GOODEY, J.B. 1963: Lab methods for work with plant and soil nematodes. Min. Ag. Fish & Food, Tech. Bull. 2.

GORDON, H.T. 1959: Minimal nutritional requirements of the German roach. Ann. NY Acad. Sci. 77: 290-351.

GOULDEN, C.H. 1962: Methods of Statistical Analysis. J. Wiley & Son, NY.

GOURLAY, E.S. 1930: Preliminary host list of the entomophagous insects of New Zealand. NZ, DSIR, Res. Bull. 22: 3-13.

HARBORNE, G.B. 1964: Biochemistry of Phenolic Compounds. Ac. Press, NY.

HARDING, J.A. 1965: Ecological and biological factors concerning the sorghum midge. Tex. Ag. Stat. Misc. Publ. 773.

HARRIS, K.M. 1966: Gall midge genera of economic importance. I. Trans. roy. Ent. Soc. Lond. 118: 313-58.

——— 1968: Systematic revision and biol. review of Cecidomyiidae predators on world Coccoidea. Trans. roy. Ent. Soc. Lond. 119: 401-94.

HATHAWAY, D.E. 1958: Evidence for the translocation of phenolics in plants. Biochem. J. 70: 34-6.

HAVILAND. 1920: On the bionomics and development of Lygocerus parasites of Aphidius. Quart. J. Micro. Sci. 65: 101-27.

HEAD, G.C. 1971: Quantitative Soil Ecology. J. Phillipson, ed. Blackwell.

HEDLIN, A.F. 1961: Life history and habits of a midge Contarinia oregonensis in Douglas fir cones. Can. Ent. 93: 952-67.

——— 1964: Life history and habits of a midge Mytophaga thujae in West Red Cedar cones. Can. Ent. 96(1): 950-1.

——— 1964: A 6 year plot study on Douglas fir cone insect population fluctuations. For. Sci. 10(1).

HEDLIN, A.F. & JOHNSON, N.E. 1963: Life history and habits of a midge Contarinia washingtonensis. Can Ent. 5(11): 1168-75.

HEDLIN, A.F. & JOHNSON, N.E. 1968: A new species of Camptomyia from Douglas fir cones. Can. Ent. 100: 532-35.

HENSON, W.R. 1959: Some effects of secondary dispersive processes on distribution. Amer. Nat. 93: 315-20.

HILLMAN, W.S. & GALSTON, S.W. 1961: The effect of external factors on auxin content. Encyl. Pl. Physiol. 14: 683-700.

HOLLOWAY, HOY *et al*. 1955: Mortality of rata/kamahi protection forests, Westland. NZFS Rpt.

HOUSE, H.L. 1969: Role of nutritional factors in forest selection and preference.....*Pseudosarophaga officinis* (Dipt.). Can Ent. 99: 1310-21.

——— 1969: Effects of different proportions of nutrients in insects. Ent. Exp. Appl. 12: 651-69.

HOY, J.M. 1958: Coccids associated with rata and kamahi in New Zealand. NZ J. Sci. 1: 179-200.

HUETTEL, M.D. & BUSH, G.L. 1972: The genetics of host selection and its bearing on sympatric speciation in *Procecidochares*. Ent. Exp. Appl. 15: 338-42.

HUGHES, J.C. & SWAIN, T. 1960: Scopolin production in potato tubers infected with *Phytophthora infestans*. Phytopath. 50: 398.

IMMS, A.D. 1957: A General Textbook of Entomology. Methuen, Lond. 886 pp.

JACKSON, M.B. & OSBORNE, D.J. 1970: Ethylene: the natural regulator of leaf abscission. Nature 225: 1019-22.

JANKIEWICZ, L.S., PLICH, H. & ANTOSZEWSKI, R. 1972: Preliminary studies on the translocation of C^{14}-labelled assimilates towards the gall of *Cynips quercus-folii*. Marcellin: 163-72.

JERMY, T., HANSON & DETHIER. 1966: Feeding inhibitors and food preference in chewing phytophagous insects. Ent. Exp. Appl. 9: 1-12.

——— 1968: Induction of specific food preferences in Lepin larvae. Ent. Exp. Appl. 11: 211-230.

JOHNSON, N.E. 1963: A new species of cecid infesting cones of Douglas fir. Ann. Ent. Soc. Amer. 56: 94-103.

JOHNSON, C.G. 1967: Migration and Dispersal of Insects by Flight. Methuen.

KAMAL, M. 1939: Biological studies on some hymenopterous parasites of aphidophagous symphidae. Egypt, Min. Ag. Tech. Bull. 207. 110 pp.

KEITH, R.W. *et al*. 1958: Quantitative paper chromatographic determination of phenols. J. Chromat. 1: 534-6.

KOZLOWSKI, T.T. 1971: Growth and Development of Trees. II. Ac. Press, NY.

KUHNELT, W. 1961: Soil Biology. Walker, N., transl.)

LAMB, K.P. 1960: A check list of New Zealand plant galls. Trans. roy. Soc. NZ 88: 121-39.

LECATSAS, G. 1966: Fine structure of insect induced plant tumour. Nature 212: 830-1.

—— 1966: Nucleonids and plant tumours. South Afr. J. Sci.: 352-4.

LEVIN, D.A. 1971: Plant phenolics: an ecological perspective. Am. Nat. 105: 157-81.

—— 1973: Plant trichomes and defence. Bio Rev.

McCALLA, D.R. 1965: Chemical nature of gall factor. Plant Physiol. 37: 98-103.

MAGNER, L. & DESSART, P. 1967: La reclassifas des catagories des Ceraphroniodea. Bull. Int. Sci. nat. Belg. 43(22): 1-33.

MANI, M.S. 1964: Ecology of Plant Galls.

MASON, R. 1958: Foods of the Australian opossum in New Zealand indigenous forest in Orongorongo Valley, Wellington. NZ J. Sci. 1: 580-613.

MASTERS, S.E., HOLLOWAY, J.T. & McKELVEY, P.J. 1957: National Forest Survey of New Zealand 1955. I. The indigenous Forest Resources of New Zealand. Government Printer, Wellington. 107 pp.

MILES, P.W. 1968: Studies on the salivary physiology of plant bugs. J. Insect Physiol. 14: 97.

—— 1969: Insect salivary enzymes and plant pathologies. Ann. Rev. Phytopath. 6: 137.

MILES, P.W. & LLOYD, I. 1967: Auxins and saliva. Aphids. Nature 213: 801.

MILNE, A. 1957: The natural control of insect populations. Can. Ent. 89: 193-213.

—— 1962: On a theory of natural control of insect populations. J. theor. Biol. 3: 19-50.

MILNE, D.L. 1961: The function of the sternal spatula in gall midges. Proc. roy. Int. Soc. Lond. A.36: 126-31.

MORRIS, R.F. 1955: The development of sampling techniques for forest insect defoliators. Can. J. Zool. 33: 225-94.

—— 1960: Sampling insect populations. Ann. Rev. Ent. 5: 243-64.

NEWCOMBE, E.H. 1951: Comparative studies of metabolism in insect galls and normal tissues, in Plant Growth Substances (F. Skoog, ed.). Uni. Wisconsin Press. 476 pp.

NICHOLSON, A.J. 1933: The balance of animal populations. J. Anim. Ecol. 2: 132-78.

——— 1957: The self adjustment of populations to change. Cold Spring Harb. Symp. quart. Biol. 22: 153-72.

NICHOLSON, A.J. & BAILEY. 1935: The balances of animal populations. Proc. Zool. Soc. Lond. 3: 551-98.

NIERENSTEIN, M. 1930: Interrelation between gall producer and gall. Nature 125: 348-9.

OGDEN, J. 1971: Studies on the vegetation of Mt Colenso, New Zealand. II. The population dynamics of red beech. Proc. NZ Ecol. Soc. 18: 66-75.

OLSEN, J.C. 1963: Gross and nett production of terrestial vegetation. J. Ecol. 52: 99-115.

PANELIUS, S. 1965: A revision of the European gall midges of the Subfamily Porriondifinae (Dipt. Itonididae). Acta. zool. fenn. 113: 1-157.

PETEL, F. <u>et al</u>. 1960: Growth <u>in vitro</u> of tissues isolated from normal stems and insect galls. Am. J. Bot. 47: 186-95.

PIMENTEL, D. 1961: On a genetic feed-back mechanism regulating popu;atopms pf herbivores parasites and predators. Am. Nat. 95: 65-79.

PITCHER, R.S. 1957: Abrasion of sternal spatula of larvae of <u>Dasyneura tetarsi</u> during post feeding stages. Proc. roy. Ent. Soc. Lond. A.32: 83-9.

PREBBLE, M.C. 1943: Sampling methods in population studies of the European spruce sawfly in Eastern Canada. Trans. roy. Soc. Can.37: 93-126.

REDMOND, D.R. 1959: Mortality of rootlets in balsam fir defoliated by the spruce budworm. For. Sci. 5: 64-9.

REFAI, MILLER, JONES & WOLFE. 1956: The feeding mechanisms of Hessian-fly (Cecid) larvae. J. Econ. Entom. 49: 182-4.

RUBIN, B.A. & ARTSIKHOVOSKAY. 1963: Biochemistry and Physiology of Plant Immunity. Pergamon Press.

SCHOONHOVEN, L.M. 1969: Amino-acid receptors in larvae of <u>Pinus brassicae</u>. Nature 221: 1268.

——— 1973: Plant recognition by Lepidopterous larvae. Symp. roy. Ent. Soc. Lond. 6: 87-99.

SEQUEIRA, L. 1965: Origin of indolacetic acid in tobacco plants infected with <u>Pseudomonas solanacearum</u>. Phytopath 55: 1232-6.

SHELDRAKE, A.R. & NORTHCOTE, D.H. 1968: The production of auxin by autolysing tissues. Planta 80: 227-36.

SLOBODKIN, L.B. 1962: Growth and Regulation of Animal Populations. Ac. Press, NY. 184 pp.

SOUTHWOOD, T.R.E. 1966. Ecological Methods. Methuen.

——— 1973: The insect/plant relationship - an evolutional perspective. Symp. roy. Ent. Soc. Lond. 6: 1-31.

SPENCER, H. 1926: Biology of the parasites and hyperparasites of aphids. Ent. Soc. Am. Ann. 19: 119-57.

STEWARD, F.C. & KRIKORIAN, A.D. 1971: Plants, Chemicals and Growth. Ac. Press, NY.

STRIDE, G.O. & STRAATMAN. 1962: The host plant relationship of an Australian swallowtail *Papilio aegeus*. Proc. Linn. Soc. NSW 87: 69-78.

SWAIN, T. & HILLIS. W.E. 1959: The phenolic constituents of *Prunus*. J. Sci. Fol. Ag. 10: 63.

TAYLOR, W.H. 1954: Soil Survey of New Zealand. NZ Soil Bureau Pub. 109.

THIELGES, B.A. 1968: Altered polyphenol metabolism in the foliage of *Pinus sylvestris* associated with European pine sawfly attack. Can. J. Bot. 46: 724-5.

THORSTEINSON, A.J. 1960: Host selection and phytophagous insects. Ann. Rev. Ent. 5: 193-218.

TORREY, J.L. 1950: Induction of lateral roots by IAA and root decapitation. Am. J. Bot. 37: 257-64.

——— 1958: Experimental modification of developmental roots, *in* Cell Organisation (D. Rudnick, *ed.*). Ronald Press, NY.

TORREY, J.L. & FOSKET, D.E. 1970: Cell division in relation to cytological differentiation in cultured pea segments. Am. J. Bot. 57: 1072-80.

UTIDA, S. 1955: Fluctuations in the interacting populations of host and parasite in relation to the biotic potential of the host. Ecology 36: 202-206.

——— 1957: population fluctuation - an experimental and theoretical approach. Cold Spring Harb. Symp. quart. Biol. 22: 139-52.

VALENTINE, E.W. 1967: A list of the hosts of ontomophagous insects of New Zealand. NZ J. Sci. 10: 1100-209.

VAN EMDEN & BASHFORD, M.P. 1971: The performance of *B.brassicae* and *M.persicae* in relation to plant age and leaf amino acid. Ent. Exp. Appl. 14: 349-60.

VARLEY, G.C. & GRADWELL, G.R. 1960: Key factors in population studies. J. Anim. Ecol. 29: 399-401.

VITE, J.P. & PITMAN, G.B. 1968: Bark beetle aggregation: effects of feeding on the release of pheromones in <u>Dendroctonus</u> and <u>Ips</u>. Nature 218: 129-70.

WALTER, E.V. 1941: The biology and control of sorghum midges. USDA Tech. Bull. 778.

WARDLE, P. 1971: Biological flora of New Zealand. <u>Metrosideros umbellata</u>, Southern Rata. NZ J. Bot. 9(4): 445-71.

WHITTAKER, R.U. & FENNY, P.P. 1971: Allelochemics: chemical interaction between species. Science, NY, 171: 757-70.

WHITTINGTON, W.J. (<u>ed.</u>) 1969: Root Growth. Butterworth, Lond. pp. 581-98.

WIGGLESWORTH, V.C. 1968: The Life of Insects. Mentor, Lond. 382 pp.

WYATT, I.J. 1963: A root infesting gall midge from New Zealand. Proc. roy. Ent. Soc. Lond. B.32: 103-7.