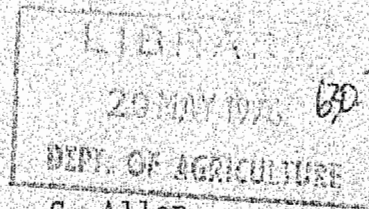


DEPARTMENT OF AGRICULTURE, SOUTH AUSTRALIA

## Agronomy Branch Report

SEQUENTIAL SAMPLING FOR THE PASTURE COCKCHAFER,  
*Aphodius tasmaniae* HOPE (COLEOPTERA: SCARABAEIDAE),  
IN PASTURES IN SOUTH AUSTRALIA.



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### 3.3.2 Pest damage assessment techniques

Various techniques to assess pest damage were outlined by Smith (1967) and have been used by other people, also. The techniques can be briefly summarised by the following, though some studies may involve more than one of these techniques -

- Comparison of damage before and after introduction of a pest into a district (Smith 1967)
- Comparison of damage before and after introduction of successful control measures into a district (Noone 1958).
- Comparison of naturally infested plants or areas with naturally uninfested plants or areas (Strickland 1954, Everly 1960).
- Cage studies in the field (Raw et al. 1957, Deane et al. 1957).
- Cage or pot studies in the laboratory or glasshouse (Davidson et al. 1968, Davidson 1969).
- Artificial infestation of pests (Brazzel et al. 1957)
- Artificial removal of pests (Judenko et al. 1952)
- Manipulation of natural enemies of pests (Thomas 1954)
- Simulated damage (Chester 1945, Kincade et al. 1970)
- Chemical treatment of pests (Franklin 1953, Joyce 1959).

The best method to assess crop damage caused by a pest depends on the pest species, type of injury and the particular crop (Judenko 1965a). The above methods are therefore discussed below to evaluate the best method of assessment of A. tasmaniae damage to pastures.

#### Comparison of damage before and after introduction of either a pest or successful control measures into a district

These two methods probably could be applied to A. tasmaniae in marginal areas of its known occurrence where it is spreading into new areas, but would give mainly an indication of the total impact of A. tasmaniae on pasture production for an area. This would be little use when attempting to evaluate losses within an individual pasture.

### Comparison of naturally infested areas with naturally uninfested areas

There are many reasons why a comparison of naturally infested areas with naturally uninfested areas may lead to biased observations on the effects of larvae. These biases apply to both intra and inter-paddock comparisons. The initial pasture composition and quality is usually different between infested, especially heavily infested, and uninfested pasture. This difference is due to the female beetle's preference to lay eggs in bare or sparsely covered areas in pasture during late summer; such areas usually having a high component of winter annual weeds.

Also, there can be a large variation in soil type and plant nutrients between paddocks and even within paddocks which will influence pasture production. If comparisons in pasture production between infested and uninfested areas are made where the pasture is grazed by livestock and unfenced, selective grazing of uninfested areas may occur. Such selective grazing becomes more apparent as feed in the infested areas becomes scarce or less favourable to livestock.

### Cage studies in the field

The use of cages to contain insects in the field makes it almost impossible to eliminate modifications of the microenvironment due to the cage. The cage must be critically designed to minimise these effects (Deane *et al.* 1957). Cages or enclosures for A. tasmaniae larvae in pasture could be made with vertical barriers set around given areas of pasture. The barriers could be made from approximately 15 cm wide galvanised metal stripping laid vertically in the ground with about 5 cm above ground level. Larvae could not escape from this type of enclosure because they move vertically in the soil and are unable to climb vertical surfaces. The size of the enclosures may limit the effectiveness of this technique. If there are high densities of larvae in relatively small areas of pasture in the enclosures, the damage may appear excessive because larvae can not disperse when food becomes less abundant, as occurs with older larvae in natural infestations. Even with moderate densities in natural, unrestricted infestations, larvae may disperse and the plants which are left and not destroyed by larvae may grow better and partly compensate for the damage as the competition between plants for light and nutrients becomes less. Also, the metal barriers could reduce the surface run-off of water during rain and increase the adverse effect of water-logged soils on larvae. The importance of this effect would vary with soil type, precipitation and topography of the site.

Cages or coverings could be used also to prevent oviposition and so provide areas without larvae which are adjacent to areas that are naturally infested; comparisons in production could then be made between the areas. The usefulness of this technique is limited by the size of the area which could be kept free of larvae.

### Cage or pot studies in the laboratory

When damage assessment studies are carried out with pasture pests in pots in a laboratory or glasshouse, it is difficult to extrapolate any differences in pasture production to "broad-acre" situations. However, this technique can be useful for determining the mode of damage and the level of damage which might be expected in the field prior to conducting field trials. With A. tasmaniae, the mode of damage is well understood and experience with this pest has given an indication of the order of losses which may be expected.

### Artificial infestations of pests

Artificial populations of A. tasmaniae larvae could be established in the field, either by caging newly-emerged adults over pasture or seeding early-instar larvae or eggs into a pasture. This method would be appropriate mainly for small plot trials which may restrict its usefulness.

### Artificial removal of pests

Artificial removal of A. tasmaniae larvae is not applicable because of the obvious destruction of the pasture with mechanical methods. Other methods of removal, e.g. electrical stimulation and the repellent effect of chemicals (Satchell 1955), would not result in absolute removal where areas free from larvae are required and, also the treatment may have other effects on the system.

### Manipulation of natural enemies of pests

Manipulation of natural enemies to give a range of pest densities is not applicable to A. tasmaniae. The main parasite is a thynnid wasp, Tachynomia sp., which has a limited distribution and is effective only within 30-40 metres of Melaleuca or Eucalyptus trees; the males feed on the blossoms of trees of these genera (Maelzer 1962a). The fungus, Cordyceps aphodii, Mathieson, influences the abundance of A. tasmaniae larvae in soils with a relatively high moisture content (Maelzer 1962a). Although development of a technique to spread spores of this fungus in a trial site may be possible, its application to damage assessment studies is limited. Infection with this fungus would not give a quick "knock-down" of larvae and would enable some feeding on green plant material to occur.

### Simulated damage

The interpretation of dry matter losses of pasture caused by artificial defoliation of pasture to simulate A. tasmaniae damage would be difficult and misleading. The main problem is that simulated damage has no concept of continuing damage which occurs naturally during the feeding stages of the larvae. Also, to express expected losses against larval densities, the quantity of pasture eaten by each larva would still have to be determined by other methods.

### Chemical treatment of pests

Insecticides have been used widely in insect damage assessment studies (Strickland et al. 1967). Usually, part of a natural pest infestation is treated with an insecticide to give almost complete control of the pest and then crop yields from this area are compared with yields from untreated areas. Also, different rates of insecticides can be used to create differences in pest densities across an infestation. However, where insecticides are used to modify pest densities, the insecticides may effect plant yields independently of pests other than the species being studied (Davies et al. 1966, Luckman 1960), or alter the effects of other pests already present (Pruess et al. 1958). While reviewing the use of insecticides in damage assessment studies, Smith (1967) concluded that the large number of factors complicating the clear interpretation of insecticidal experiments almost precluded their use in establishing the impact of insects on crop yields. Strickland et al. (1967) were not as unyielding in their criticism but warned that the use of insecticides can not give true estimates of losses from specific pests unless the side effects are known to be unimportant. In contrast to both Smith and Strickland et al., Le Clerg (1971) relied heavily on paired-treatment experiments using insecticides when he outlined experimental techniques to assess losses caused by insects. Le Clerg's techniques were included in the FAO manual on "Crop Loss Assessment Methods" which was designed to provide principles on which to base the planning and conducting of field experiments to measure crop losses.

After considering the above methods, and their possible combinations, to assess pest damage, the most appropriate method to assess A. tasmaniae damage to pasture appears to be the surface application of insecticide to provide areas practically free of larvae in naturally infested pastures. Comparison of pasture production from these areas can be made with production from adjacent areas where larval densities have been estimated. Any differences in pasture production can be attributed mainly to larval activity. There is no information on the effect of surface applications of lindane on pasture growth. However, any effects caused by the low rate of application (280 g a ha active ingredient) necessary to give practically complete control of larvae would probably be insignificant compared to the expected damage caused by the larvae. A. tasmaniae usually occurs as the only pest species when it infests pasture in South Australia, however, during autumn and spring, H. destructor and S. viridis may also infest the same pasture. If the latter two pests need to be controlled in a trial, a surface spray of phosmet will give good control and should not greatly affect the A. tasmaniae larvae nor pasture growth because only a very low rate of application (50 g a ha active ingredient phosmet) is required. Low densities of other scarab larvae can be found in A. tasmaniae infested pastures but these scarabs do not come to the soil surface and would be unaffected by the surface application of lindane. There are no known situations where

treatment for A. tasmaniae has led to an increase in either the density of another pest or the stimulation of another species to pest status.

#### 4. OBSERVATIONS ON THE DISTRIBUTION OF A. tasmaniae LARVAE

The frequency distributions of natural populations of early-instar A. tasmaniae larvae in pastures were estimated during a three-year period. The pastures mainly consisted of annual plant species, though some had a minor component of perennial species.

Stratified random sampling (by area) was used to estimate the distributions of all populations sampled. This method of sampling was preferred to random sampling because it was more likely to give any individual in the population an equal chance to be sampled (Healy 1962, Yates et al. 1942). The population being sampled was divided into the required number of equal-sized strata and one sample unit was randomly taken from each stratum. The corresponding corner of each stratum was located on a grid pattern marked out in the field by two sets of pegs placed at right angles to each other. Each set had two parallel rows of equally spaced pegs. From these corners, random numbers were used to define the exact position of the sample unit; this avoided biased sampling which might occur due to soil casts on the soil surface indicating the presence of larvae. The sample unit size and number of sample units taken varied from year to year and depended on the previous year's results. Each sample unit was all the soil within the sample unit area, defined precisely by the area of a corer, to a depth which included all larvae within that area. The depth varied with the stage of growth of the larvae and soil type. The soil was transferred to a plastic bag, sealed and taken to the laboratory for hand sorting of larvae.

##### 4.1 Larval Distributions - 1970

###### 4.1.1 Sampling areas

Sampling was restricted to populations of larvae in pastures based on annual ryegrass and subterranean clover. Populations were chosen to give a range of mean larval densities and other ecological factors which could influence the sampling distribution of larvae (see Section 3.2). To ensure that the populations did differ in density, quick estimates of densities were obtained along a number of transects across the area occupied by the population. The area sampled within a paddock varied between two to four hectares.

Populations were sampled at Nairne, Harrogate and Meadows in the Adelaide Hills and at Pareena and Wye in the Lower South-East of South Australia.

Table 1, describes, for 1970, the time of sampling, the prevalent instars at sampling and a description of the sample areas.

###### 4.1.2 Frequency distributions and larval densities

Each area of infestation was divided into 100 equal strata and a sample unit, 400 cm<sup>2</sup>, was taken randomly from each stratum.

The mean number and range of larvae per sample unit for each population sampled are shown in Table 2.

The frequency distribution of larvae for each population is shown in Figure 3.

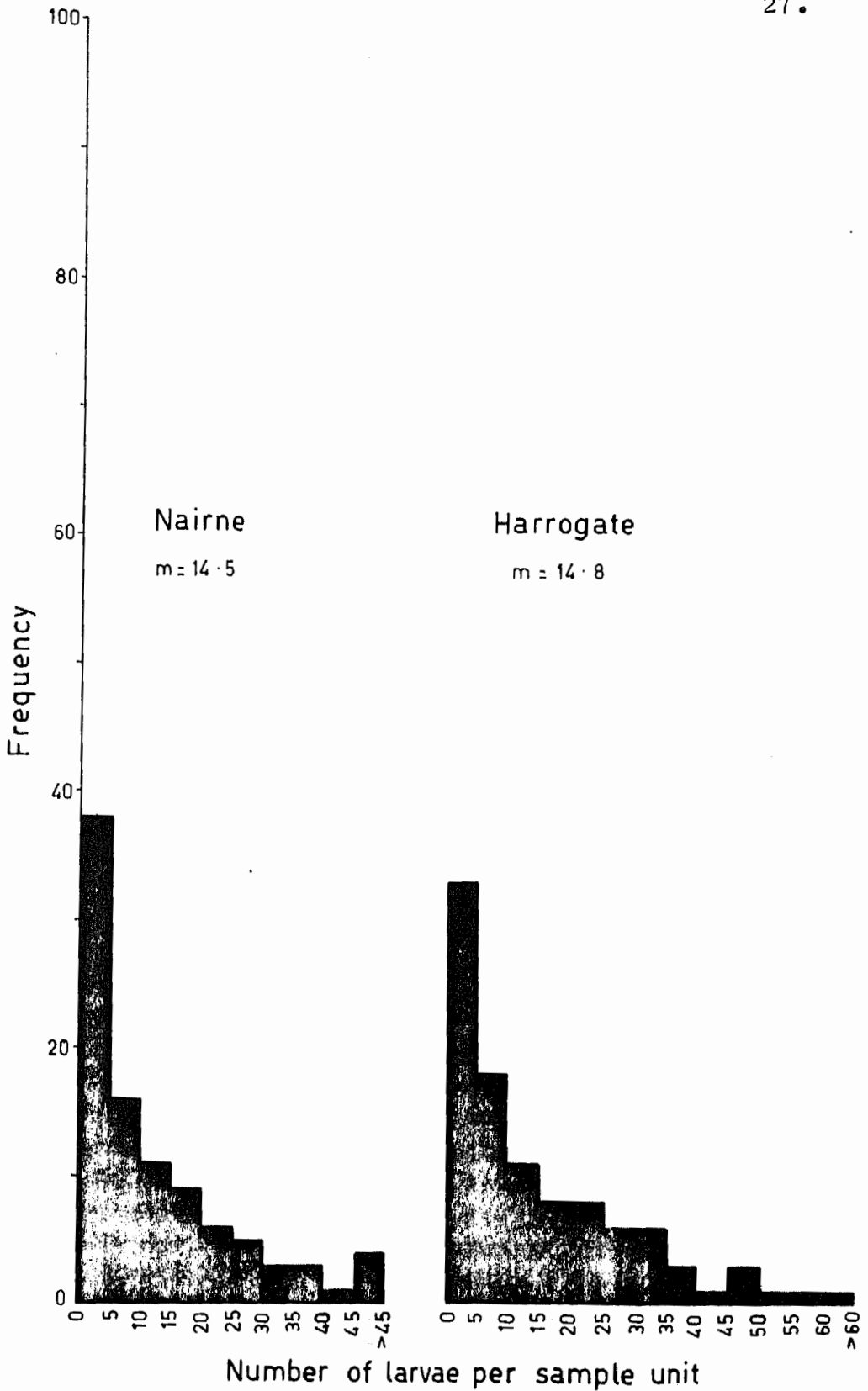
Table 1. Time of sampling, the prevalent larval instars at sampling and a description of the sample areas, 1970.

Location	Nairne	Harrogate	Meadows	Pareena	Wye
Date of sampling	27/4/70	5/5/70	21/5/70	23/6/70	30/6/70
Larval instar (s)	early second	mid-second, early third	early third	early third	late second, early third
Average annual rainfall (mm)*	690	760	890	690	710
Pasture species	Annual ryegrass, subtterranean clover, capeweed, crowfoot	Annual ryegrass, subtterranean clover, capeweed, crowfoot	Annual ryegrass, subtterranean clover, capeweed, crowfoot	Annual ryegrass, subtterranean clover, capeweed, crowfoot	Annual ryegrass, subtterranean clover, capeweed, crowfoot
Pasture quality	Poor	Poor	Poor	Fair	Poor
Soil type	Red podsollic	Podsollic	Meadow podsollic	Terra-rossa	Terra-rossa
Topography	North and south facing slopes - gully down centre	South facing slope	Flat	Slight north facing slope	Flat
Presence of trees (5-20m)	-	+	+	-	-
Previous <i>A. tasmanica</i> activity	1969	1968 - light 1969 - treated	-	1969	-

\* Estimated from "Rainfall Statistics South Australia", Director of Meteorology, Melbourne, 1966.







m - mean number of larvae per sample unit

Figure 3 : Frequency distributions of larvae in populations of *A. tasmaniae* sampled with a 400 cm<sup>2</sup> sample unit, 1970.  
(Cont'd.)

Table 2. Mean number and range of larvae per sample unit for each population sampled in 1970.

Location	Mean (No. of larvae per sample unit)	Range (No. of larvae per sample unit)	Mean (No. of larvae per m <sup>2</sup> )
Harrogate	14.8	0-75	370
Nairne	14.5	0-104	362
Pareena	11.9	0-42	298
Meadows	7.5	0-36	188
Wye	2.9	0-18	72

#### 4.1.3 Discussion

Sampling with a 400 cm<sup>2</sup> sample unit did not provide frequency distributions which could generally be fitted to negative binomial distributions nor any other distribution which could be applied to a sequential plan; the distributions were too positively skewed. The distribution which most closely approached a negative binomial distribution occurred with the infestation which had the lowest mean number and range of larvae per sample unit. This suggested that reducing the size of the sample unit would ensure a smaller range of larvae per sample unit and could provide distributions more closely approaching negative binomial distributions.

In addition to improving the chance for the distributions to fit a negative binomial distribution, reducing the sample unit area had two other advantages. Firstly, sequential plans based on a smaller sample unit would have better acceptance by farmers and agricultural advisers because of the smaller quantities of soil which would need sorting in the use of the plan. Secondly, if a smaller sample unit did not provide distributions which could be used in a sequential plan, sampling units could be summed to give larger units that would be expected - by the Central Limit Theorem (Clarke 1969) - to approximate to normal distributions. Application of the Central Limit Theorem involves taking a series of samples of  $n$  sample units from the parent distribution and plotting a frequency distribution of the total number of larvae for each larger sample of units. Parameters of the resultant normal distributions can then be used in a sequential plan. It should be noted that the number of sample units on the abscissa of a sequential plan would become the number of samples, where each sample would contain an equal number of sample units. To maintain the practicability of a sequential plan, the number of sample units required for each sample to approximate a normal distribution would have to be low. When the Central Limit Theorem was applied to data of the distributions in Figure 3, only five sample units from the sampling at Wye needed to be summed to give a new distribution which approximated to a normal distribution.

At least ten sample units had to be summed when the data from the other distributions, which had higher mean densities and ranges of larvae per sample unit than the data from Wye, were used. Hence, using a smaller sample unit, as suggested above, would restrict the mean number and range of larvae per sample unit and probably provide data that required a minimum number of sample units to be summed to give a new distribution which could be approximated to a normal distribution. This procedure would only be necessary if the use of a smaller sample unit did not provide a distribution which could be described by a negative binomial distribution or any other distributions which could be used in a sequential plan.

Reduction of the sample unit to  $108 \text{ cm}^2$  (11.7 cm (4") diameter core) appeared to be a useful size, especially when application of the plan was considered, because a soil corer or post-hole digger with this diameter is found on most properties.

The frequency distributions in Figure 3 indicated a relationship between mean larval density and the distribution, regardless of differences in other ecological factors between the sampling areas which might have influenced the distribution of larvae. This apparent relationship between the mean larval density and the distribution suggested that parameters of distributions of populations used in a sequential plan must come from distributions describing those populations which have a mean larval density approximating the economic injury level used in the plan.

#### 4.2 Larval Distribution - 1971

##### 4.2.1 Sampling area

In 1971, most of the available time was committed to damage assessment studies, but one population was sampled to test whether a  $108 \text{ cm}^2$  sample unit would provide a frequency distribution which could be fitted to a negative binomial distribution. Because of the apparent relationship between mean larval density and the sampling distribution in 1970, an infestation was selected where the mean larval density was estimated with a spade, as in Section 4.1.1., to be approximately 150 larvae per  $\text{m}^2$ . This level of infestation was chosen because it was a density of larvae in pasture which has been considered for many years to be "worth" treating in a "normal" year. The value was derived from experience, not experimentation, but provided a guide in initial studies.

The population was at Moorak in the Lower South-East of South Australia. Table 3 describes the time of sampling, the prevalent larval instars at sampling and a description of the sample area.

Table 3. Time of sampling, the prevalent larval instars at sampling and a description of the sample area at Moorak, 1971.

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Date of Sampling	5/7/71
Larval instars	early to mid-third
Average annual rainfall (mm)*	790
Pasture species	Annual ryegrass, subterranean clover, capeweed.
Pasture quality	Poor, thinning of pasture due to <u>A. tasmaniae</u> becoming evident.
Soil type	Sandy loam with patches red-clay over limestone.
Topography	Flat
Presence of trees (5 - 20m)	-
Previous <u>A. tasmaniae</u> activity	1970

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\*Estimated from "Rainfall Statistics South Australia" Director of Meterology, Melbourne, 1966.

#### 4.2.2 Frequency distribution and larval density

The area of infestation was divided into 150 equal strata and a sample unit, 108 cm<sup>2</sup>, was taken randomly from each stratum.

The mean number of larvae (including the range) was 1.59 (0-14) larvae per sample unit. This was equivalent to a mean density of 147 (0-1300) larvae per m<sup>2</sup>.

The observed frequency distribution of larvae and a negative binomial distribution with the same values for the parameters, m and k, respectively, are compared in Figure 4.

The expected frequencies of the negative binomial distribution were calculated using the equations -

$$E(f_0) = \frac{N}{(1 + m/k)} k$$

and

$$E(f_x) = \frac{(k + x - 1)}{x} \binom{m}{k + m} E(f_{x-1})$$

The value of  $k$  was the maximum likelihood estimate computed from the observed frequency distribution using an iterative procedure (Bliss et al. 1953);  $k$  was estimated to be 0.70.

The agreement between the two distributions was tested using -

$$\chi^2_{n-2}$$

#### 4.2.3 Discussion

The close agreement of the theoretical to the observed distribution showed that sampling with a 108 cm<sup>2</sup> sample unit provided a distribution of early instar larvae which could be fitted to a negative binomial distribution when the density was about the expected economic injury level. Parameters of this distribution could not be used for the basis of a sequential plan until it was known whether or how the density of larvae and other ecological factors influenced the distribution, especially the value of  $k$ .

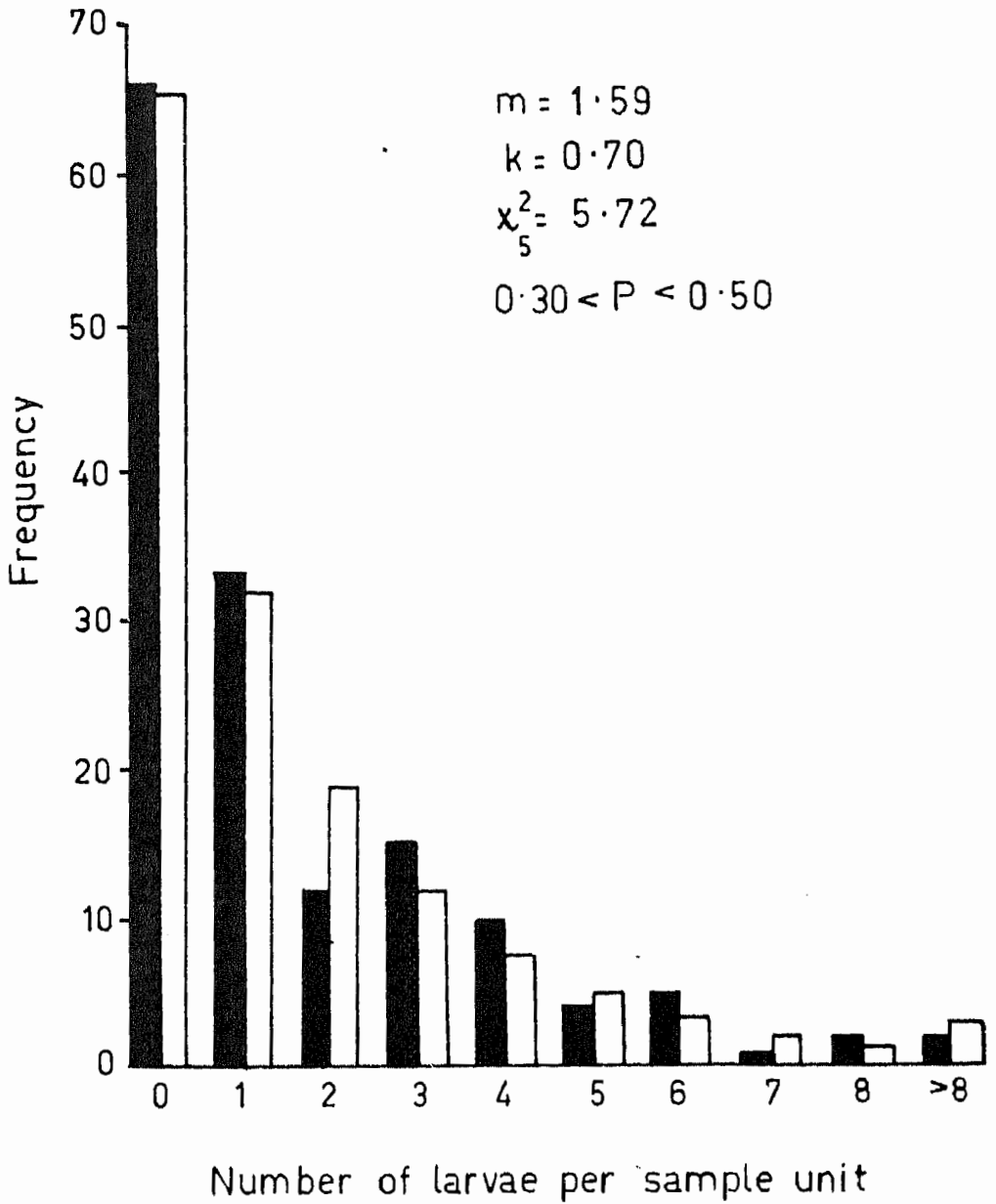
Initially, any effects could be estimated only by sampling a number of infestations in pastures which had different densities of larvae and occurred in paddocks with different combinations of other ecological factors which may influence the distribution (see Section 3.2).

### 4.3 Larval Distributions - 1972

#### 4.3.1 Sampling areas

Five infested pastures were sampled with a 108 cm<sup>2</sup> sample unit. The populations were chosen to give a range of densities of larvae up to approximately 200 larvae per m<sup>2</sup>. The economic injury level for most populations in most years was expected to be within this range of densities. Also, areas of infestation were chosen to include paddocks with different combinations of ecological factors, referred to previously, which may affect the distribution of larvae. The area of infestation sampled within a paddock varied between two to four hectares.

Populations were sampled at Prospect Hill, Blackfellow's Creek, Bugle Ranges, Nairne and Birdwood in the Adelaide Hills.



m - mean number of larvae per sample unit

k - dispersion parameter of the negative binomial distribution

■ observed distribution

□ negative binomial distribution

Figure 4 : Frequency distribution of larvae in a population of *A. tasmaniae* sampled with a 108 cm<sup>2</sup> sample unit at Moorak, 1971, compared to a negative binomial distribution.

Because the results from these paddocks indicated a trend between mean density and the value of  $k$ , a further area of infestation was sampled later in the season where it was obvious that part, but not necessarily all, of the paddock required treatment. This area of infestation was sampled to test the reliability of the relationship between mean density and the value of  $k$  in populations where there were obvious patches of larvae requiring treatment and the mean density was above the expected range of densities of the economic injury level. The paddock was at Macclesfield in the Adelaide Hills.

Table 4 describes, for 1972, the time of sampling, the prevalent larval instars at sampling and a description of the sample areas.

#### 4.3.2 Frequency distributions and larval densities

Each area of infestation was divided into 200 equal strata and a sample unit,  $108 \text{ cm}^2$ , was taken randomly from each stratum.

The mean number and range of larvae per sample unit for each population sampled are shown in Table 5.

The observed frequency distributions of larvae and negative binomial distributions with the same values for the parameters,  $m$  and  $k$ , respectively, are compared in Figure 5, for each infestation.

The methods to estimate the value of  $k$  from the observed frequency distributions and the calculation of the expected frequencies of the negative binomial distributions were the same as in Section 4.2.2.

Table 5. Mean number and range of larvae per sample unit for each population sampled in 1972.

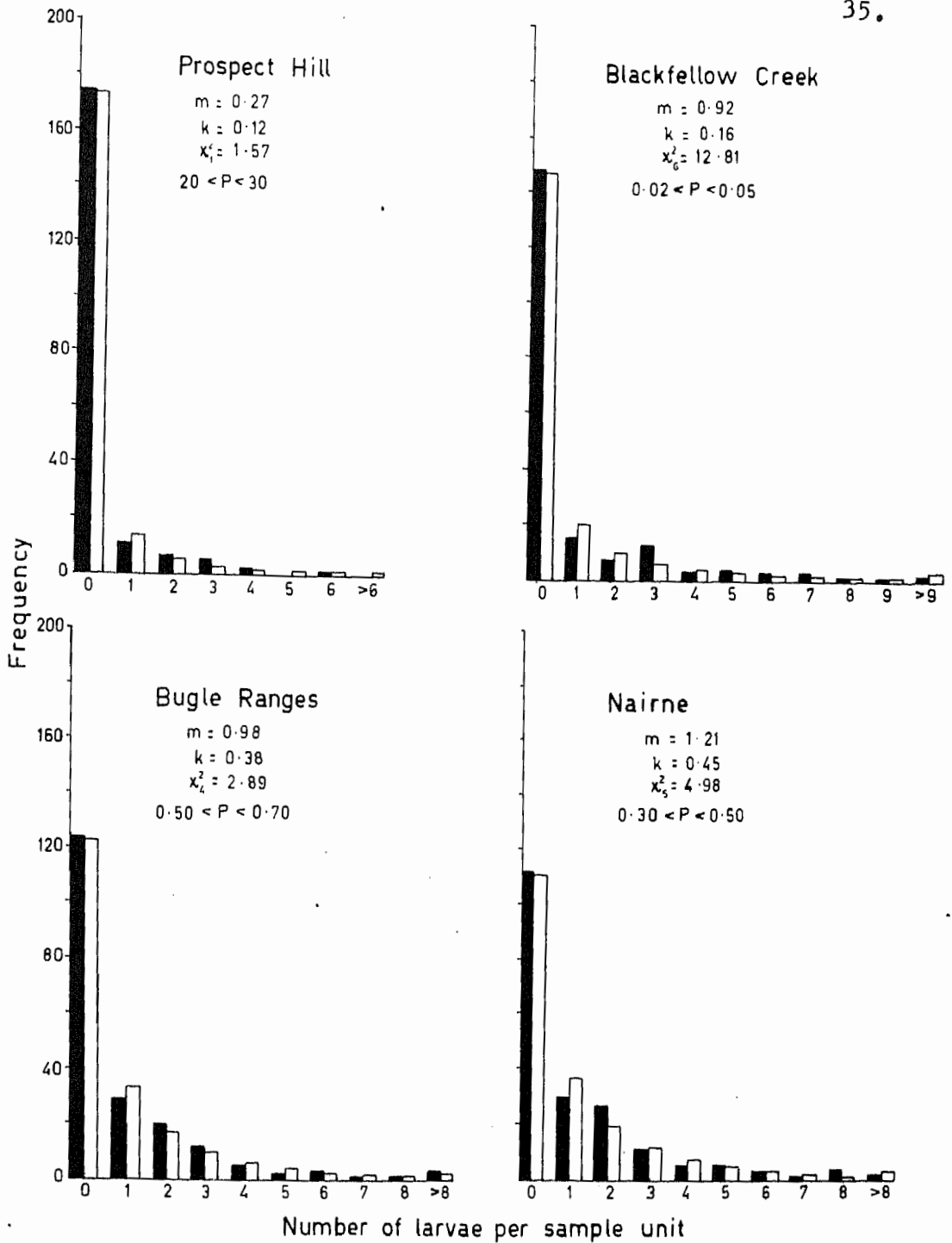
Location	Mean (No. larvae per sample unit)	Range (No. larvae per sample unit)	Mean (No. larvae per $\text{m}^2$ )
Prospect Hill	0.27	0 - 6	25
Blackfellows Creek	0.92	0 - 14	85
Bugle Ranges	0.98	0 - 10	91
Nairne	1.21	0 - 12	112
Birdwood	2.04	0 - 10	189
Macclesfield	2.98	0 - 20	277

Table 4. Time of sampling, the prevalent larval instars at sampling and a description of the sample areas, 1972.

Location	Prospect Hill	Blackfellows Creek	Bugle Ranges	Nairne	Birdwood	Macclesfield
Date of sampling	30/5/72	5/6/72	13/6/72	4/7/72	19/7/72	2/8/72
Larval instar (s)	mid-second	mid-second, late second	late second, early third	early-third	third	third
Average annual rainfall (mm)*	870	870	725	660	825	700
Pasture species	Perennial ryegrass, subtterranean clover, crowfoot	Annual ryegrass, subtterranean clover, barley grass.	Perennial ryegrass, annual ryegrass, subtterranean clover, crowfoot, cape-weed	Perennial ryegrass, barley grass	Annual ryegrass, subtterranean clover, capeweed, crowfoot	Annual ryegrass, subtterranean clover, barley grass, capeweed
Pasture quality	Poor	Good cover	Heavily grazed	Good cover	Poor	Medium - poor. Bare patches apparent in heavily infested areas.
Soil type	Meadow podsollic	Sandy podsollic	Yellow podsollic	Red podsollic	Yellow podsollic	Yellow podsollic
Topography	Flat	N.W. facing slope	West facing slope	North facing slope	North facing slope	West facing slope
Presence of trees (5-20m)	+	-	-	+	-	-
Previous <i>A. tasmaniæ</i> activity	1971 - light	1971 - light	1971 - medium	1971 - treated	1971	-

\* Estimated from "Rainfall Statistics South Australia", Director of Meteorology, Melbourne, 1966.





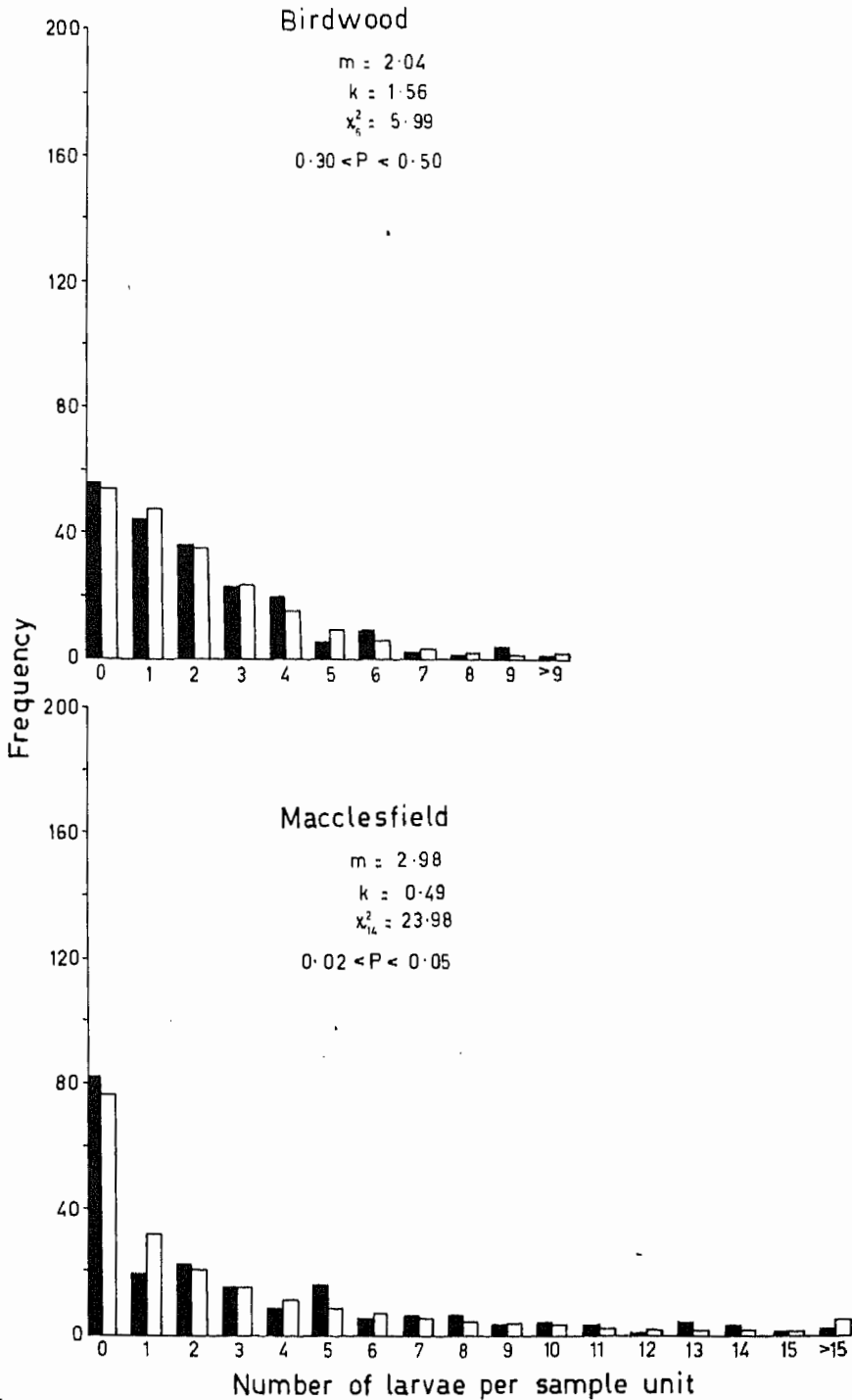
$m$  - mean number of larvae per sample unit

$k$  - dispersion parameter of the negative binomial distribution

■ observed distribution

□ negative binomial distribution

Figure 5 : Frequency distributions of larvae in populations of *A. tasmaniae* sampled with a 108 cm<sup>2</sup> sample unit, 1972, compared to negative binomial distributions.



$m$  - mean number of larvae per sample unit

$k$  - dispersion parameter of the negative binomial distribution

■ observed distribution

□ negative binomial distribution

Figure 5 : Frequency distributions of larvae in populations of *A. tasmaniae* sampled with a 108 cm<sup>2</sup> sample unit, 1972, compared to negative binomial distributions.  
 (Cont'd.)

The agreement between the expected negative binomial distribution and the observed distribution in each case was tested using  $\chi^2$ . With most distributions a number of the lower frequency<sup>n-2</sup> classes had to be grouped which explains the varying number of degrees of freedom in the  $\chi^2$ . There was an apparent relationship between the value of k and the mean number of larvae per sample unit for the different populations (Figure 6). For the purpose of the sequential plan, this was assumed to be causal and a significant linear regression was obtained if log k was plotted against mean number of larvae per sample unit (Figure 7). The regression was described by the equation:

$$\log k = -1.16 + 0.65 m, (r = 0.96)$$

The value of k estimated from data from the population at Macclesfield was not included for reasons discussed in the next section.

#### 4.3.3 Discussion

The 1972 sampling supported the 1971 data and showed that frequency distributions of populations of early-instar A. tasmaniae larvae sampled with a 108 cm<sup>2</sup> sample unit can be described by negative binomial distributions. This applied to populations where the mean density was not greater than about 200 larvae per square meter. The probability of the distribution of the population at Blackfellows Creek fitting a negative binomial distribution was very low ( $0.02 < P < 0.05$ ) mainly due to a relatively large number of sample units containing three larvae in the observed distribution. There was no apparent reason for this and it was considered to be a chance occurrence because the remainder of the observed and negative binomial frequencies were similar.

In 1972, larval sampling was carried out later in the year than normally expected due to the rainfall pattern following the early-April rains which stimulated newly-hatched larvae to come to the surface to feed. (Figure 8). The dry period from April to June retarded larval development. While some movement of larvae may have occurred in the later-sampled populations, there was little evidence of migration and food was plentiful across the areas of infestation.

The ecological factors which might affect the distribution of larvae within the infested paddocks differed markedly (Table 4), but the only factor which appeared to be correlated with the value of k, was mean larval density. The value of k increased as density increased up to a mean density of about 200 larvae per square meter. The value of k required for use in a sequential plan will vary depending on the economic injury level and can be estimated from the equation,  $\log k = -1.16 + 0.65 m$ , where m is the required economic injury level for a given situation. The relationship between k and mean density in this regression equation resulted from sampling data from populations in different parts of the Adelaide Hills, different parts of the State and from different years.

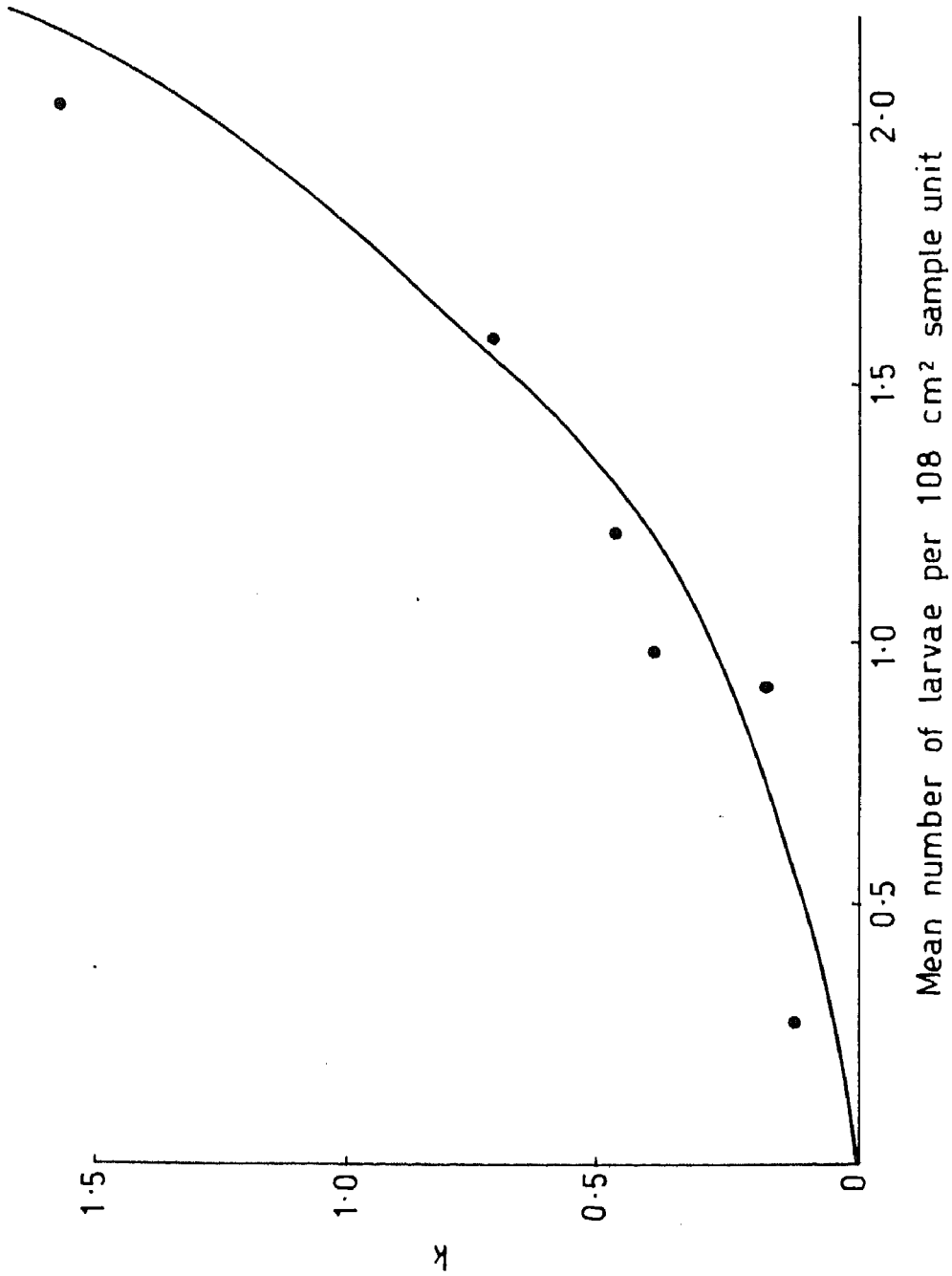
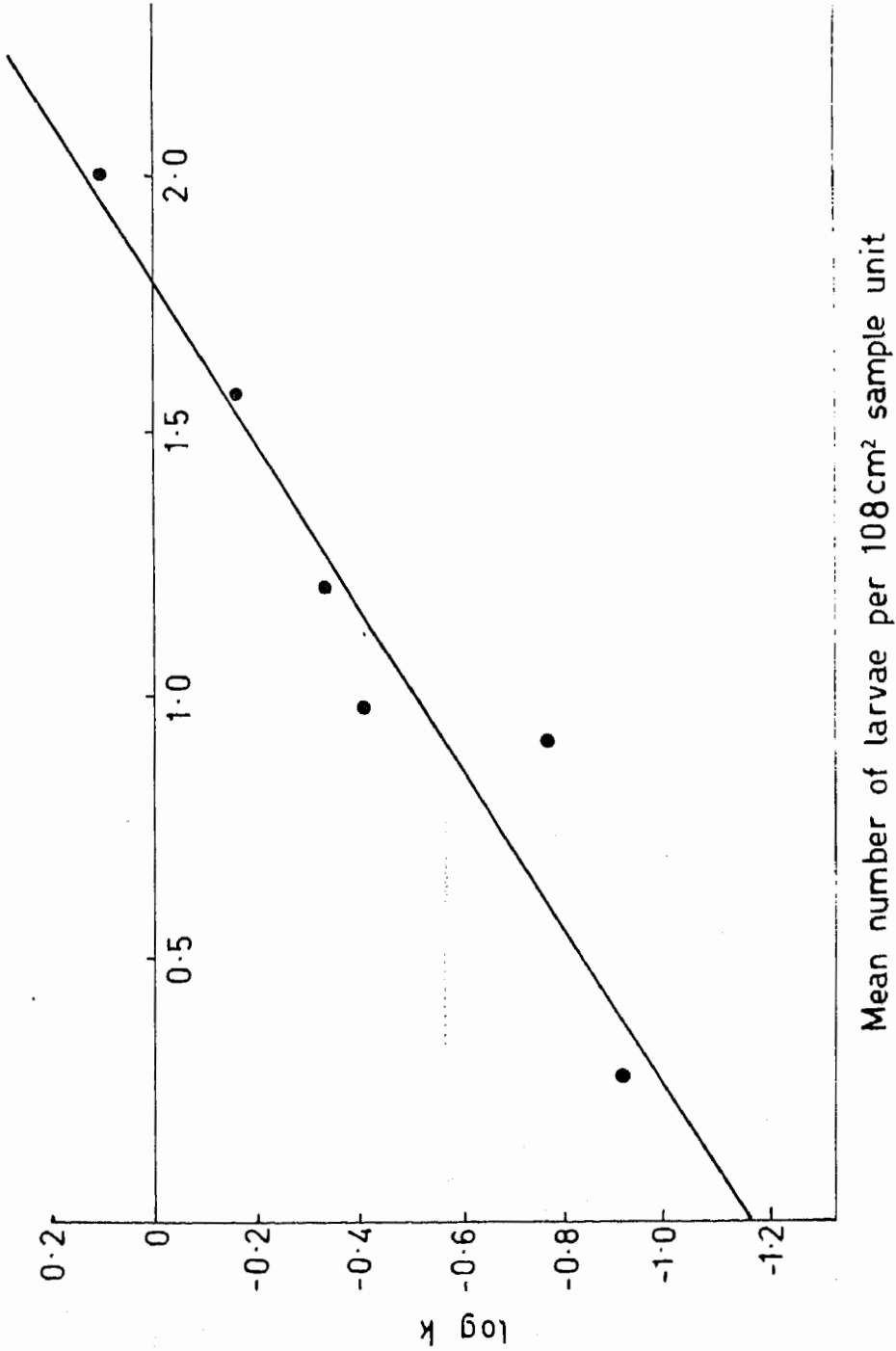


Figure 6 : Value of  $k$  plotted against mean number of larvae per sample unit for populations of *A. tasmanicae* sampled in 1971 and 1972.



Mean number of larvae per 108 cm<sup>2</sup> sample unit

Figure 7 : Value of log k plotted against mean number of larvae per sample unit for populations of *A. tasmanicae* sampled in 1971 and 1972.

The value of  $k$  for the population at Macclesfield was not included in the regression analysis because it was much lower in relation to the mean number of larvae per sample unit compared to other populations. This was partly expected because of the concentration of larvae on an old sheep "camp" which would have provided adults with a more favourable oviposition site compared to the rest of the paddock. This anomolous situation does not reduce the effectiveness of a sequential plan based on values of  $k$  estimated from populations with lower mean densities. Where there are obviously heavy concentrations of larvae in paddocks, a decision on the need for treatment of these heavily infested areas does not require a sequential plan. However, the plan may be used to make a decision with the remainder of the paddock where the need for treatment may not be as obvious.

## 5. TRIALS ON DAMAGE ASSESSMENT OF *A. tasmaniae* LARVAE

### 5.1 Ungrazed Trials

In 1971, an unreplicated, paired-treatment trial was conducted at each of four different sites to study the effect of *A. tasmaniae* larvae on the late-autumn, winter and spring dry matter (D.M.) production of annual rye-grass/subterranean clover pastures ungrazed by livestock. The two treatments were pasture naturally infested with larvae and pasture treated with lindane to control larvae, respectively.

#### 5.1.1 Methods

##### Trial sites

Four sites were selected in the Adelaide Hills in early autumn to give sites with different mean larval densities but comparable pasture composition and quality. Each site measured 40 m x 40 m and was fenced to exclude grazing livestock. Half of each site was sprayed with lindane (280 g a ha active ingredient) to control *A. tasmaniae* larvae before they began feeding on green plant material. The lindane was applied with a hand boom spray pressurised by a cylinder of carbon dioxide.

##### Larval density

The mean number of larvae in the untreated half of each site was estimated at the beginning of the trial. Fifty sample units, each a soil core 20 cm x 20 cm, were taken from each site using stratified random sampling (by area). The larvae were hand sorted from the soil in the sample units.

##### Fertilizer application

All sites, except Site II, were topdressed with superphosphate by the landowners prior to the trials. Poor subterranean clover growth in Site II after the trial started indicated phosphorus deficiency and both treatments in the trial were topdressed with superphosphate (125 kg a ha) on 13/7/71.

### Pasture production

D.M. pasture yields of both treatments at each site were estimated 6, 22 and 31 weeks after the application of lindane. Yields were estimated also at Site IV after 15 weeks.

At the six-weeks period, pasture yield was estimated by cutting pasture with a Howard forage harvester from five quadrats, each 15 m x 0.8 m, at random from each treatment. The pasture was cut to within about one centimeter of the soil surface. The pasture cut from each quadrat was weighed in the field immediately after cutting. A sub-sample of approximately 2 kg of pasture from each quadrat was placed in a plastic bag and taken to the laboratory for pasture D.M. determinations. Pasture D.M. per quadrat was determined from the percentage D.M. of the pasture in the sub-samples. The percentage D.M. was estimated by drying a known weight of fresh pasture from each sub-sample to a constant weight in an oven at 90°C.

Estimates of pasture yields using the above method of sampling were variable (the coefficient of variation ranged between 0.09 and 0.86) which made it difficult to show any significant differences between production from the treatments. For this reason, an electronic pasture probe (capacitance probe) was used to estimate pasture yields after 15 and 23 weeks. Electronic pasture probes enable rapid, non-destructive estimates of the yield of growing pasture (Alcock *et al.* 1967). Rapid sampling means that a large number of observations can be made in reasonable time which gives a more reliable estimate of the yield compared to estimates made from a few observations. The electronic pasture probe measures the change in capacitance between the probes (electrodes) of the instrument when the probes are placed in pasture. The change in capacitance varies with the moisture content of the herbage and is recorded on a meter. An electronic pasture probe can only be used confidently to estimate D.M. yields of pasture if the linear regression between the meter reading and the pasture D.M. is significant. The relationship varies markedly with different plant species and atmospheric and pasture moisture levels at the time of sampling.

The reliability of an electronic pasture probe to estimate yields of a mixed annual rye-grass/subterranean clover sward was tested prior to its use in the trial sites (Appendix II). Estimates of pasture yields in the trials using an electronic pasture probe were made by taking two hundred meter readings on a grid pattern in each treatment. The probe was calibrated for each treatment immediately after the meter readings were taken for that treatment (see Appendix II for method of calibration). Calibrating the instrument after each treatment had been sampled minimised the chance for error in the calibration due to a change in moisture which occurs during the day.

After 31 weeks, an electronic pasture probe could not be used to reliably estimate the yield of pasture because the pasture was beginning to "dry-off". Pasture D.M. yields were estimated by cutting all the herbage with hand shears from ten quadrats, each 0.8 m x 0.8 m, selected at random in each treatment and drying the pasture to a constant weight in ovens at 90°C to calculate percentage D.M.

### Mowing the trial sites

The pasture in both treatments at each site was mown to within about one centimeter of the soil surface following the estimate of pasture yields six weeks after the application of lindane. The plots were mown in an attempt to partly simulate the effect of grazing livestock; it was visually apparent that there were large differences between pasture production in the trial site compared to production from adjacent areas of the same pasture grazed by livestock and where A. tasmaniae densities were similar (see Section 5.1.3.).

### 5.1.2 Results

#### Trial sites

Table 6. Location, mean annual rainfall, pasture composition at the beginning of the trials and date of lindane treatment for the four trial sites.

	Site			
	I	II	III	IV
Location	Flaxley	Bugle Ranges	Harrogate	Woodside
Mean annual rainfall (mm)	850	700	650	720
Pasture composition - dominant spp.	Annual ryegrass, subt. clover*	Annual ryegrass, subt. clover*	Annual ryegrass, subt. clover*	Annual ryegrass, subt. clover*
- minor spp.	Capeweed	Crowfoot	Crowfoot	Capeweed
Date lindane treatment	12/5/71	7/5/71	7/5/71	12/5/71

\*Cultivar Mt. Barker



Larval densities

Table 7. Mean density and range of larvae per sample unit (400 cm<sup>2</sup>) and mean density of larvae per square meter in the untreated pasture at the beginning of the trials.

	Site			
	I	II	III	IV
Mean number of larvae per sample unit	2.6	10.4	10.8	21.7
Range of larvae per sample unit	0-10	0-49	0-72	0-69
Mean number of larvae per m <sup>2</sup>	66	259	270	542

Table 8. Summary of D.M. pasture yields and comparison of the D.M. yields between lindane treated and untreated pasture at the four sites during the trials.

Site	Weeks after treatment with lindane	Mean D.M. pasture yield (kg per ha)		$t_{n_1+n_2-2}$	Significance
		Treated	Untreated		
I	6	580 $\pm$ 24	530 $\pm$ 41	$t_8 = 1.06$	NS
	22	2870 $\pm$ 25	2860 $\pm$ 22	$t_{398} = 0.25$	NS
	31	6120 $\pm$ 210	6180 $\pm$ 280	$t_{18} = 0.18$	NS
II	6	120 $\pm$ 31	430 $\pm$ 70	$t_8 = 7.12$	***
III	6	390 $\pm$ 150	310 $\pm$ 57	$t_8 = 0.80$	NS
	22	4180 $\pm$ 41	4740 $\pm$ 34	$t_{398} = 10.60$	***
	31	6350 $\pm$ 205	6350 $\pm$ 250	$t_{18} = 0.05$	NS
IV	6	970 $\pm$ 40	710 $\pm$ 140	$t_8 = 1.72$	NS
	15	2630 $\pm$ 20	2530 $\pm$ 44	$t_8 = 2.14$	*
	22	2490 $\pm$ 23	3020 $\pm$ 47	$t_{398} = 10.20$	***
	31	7170 $\pm$ 320	6690 $\pm$ 430	$t_{18} = 0.90$	NS