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# ASPECTS OF THE COMPARATIVE BIOLOGY OF THREE WEEDY SPECIES OF AMARANTHUS IN SOUTHWESTERN ONTARIO

bу

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Voc. I

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Faculty of Graduate Studies

The University of Western Ontario

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#### ABSTRACT

The economic cost to North American agriculture of weeds is greater than the costs of all other kinds of pests. Modern methods of chemical weed control may introduce risk of concomitant environmental damage which in the long run may threaten crop yields themselves.

Knowledge of the ecological characteristics of weed species may suggest alternative methods of control that provide greater overall benefits. The purpose of this work was to gain further insights into the ecology of three important weeds and to relate this knowledge to the success of these species in the agricultural environment.

Amaranthus hybridus L., A. powellii S. Wats., and

A. retroflexus L., each of which is colloquially known as redroot pigweed. Plants of the three species are morphologically similar and a number of characters must be considered in combination if specimens are to be assigned to the correct species. Discontinuous variation is plainly seen within

A. powellii in morphological and physiological characteristics.

Amaranthus retroflexus is the most frequent species. This species and A. powellii are found in a wide range of arable habitats. Amaranthus hybridus is the least frequent species and it appears to be restricted to light-textured soils.

The seeds of each species exhibit a dormancy mechanism whereby they require a higher minimum temperature for germination when freshly harvested than after wintering in the field. The difference between the temperatures required is greatest for seeds of A. powellii and least for seeds of A. hybridus. Seeds of A. powellii that have after-ripened (lost their dormancy) germinate more rapidly at alternating temperatures of 25°/10°C than after-ripened seeds of the other species. Few seeds of A. hybridus germinate at temperatures as low as these. These characteristics can be seen as adaptations to climatic conditions. Differences between species in characteristics of seed dormancy can be invoked to explain differences in distribution.

The degree of after-ripening shown by seeds that have passed the winter under natural conditions is influenced by several factors. Seeds that have matured early in the season are more fully after-ripened than seeds that have matured late in the season. Seeds that have matured on younger plants are more fully after-ripened than seeds that have matured on older plants. Seeds that have passed the winter on or below the soil surface are more fully after-ripened than seeds that have passed the winter above the soil on plant remains. In addition, seeds of the different taxa differ in the rate at which they after-ripen. For these reasons there will usually be seeds at different stages of after-ripeneing when conditions in the spring become favourable for the germination of any seed. Furthermore, individual seeds will

exist in a variety of microenvironments under a range of temperature and light conditions. These factors influence the timing of germination of seeds of these species. Thus, there are reasons to expect that the germination of seeds in the field will be intermittent and this may prove to be advantageous to the species in withstanding weed control practices.

Knowledge of the germination behaviour of seeds of these species is discussed in relation to the observations made of the distribution and frequency of each species.

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#### CHAPTER 1

#### INTRODUCTION

#### The need for research in the ecology of weeds

The current economic losses to agriculture in the United States are approximately \$11 billion each year (Klingman 1961) of which about one-third, or \$3.7 billion, are the consequence of weeds. Losses caused by weeds are of two basic types; the costs of controlling or eradicating weeds, and the costs (in crop yield etc.) of tolerating weeds. The cost of controlling weeds in the United States is currently about \$1.5 billion a year which is over three and a half times the cost of controlling insect pests and over twelve times the cost of controlling plant diseases (Klingman 1961).

The situation in Canada would appear to be similar. In 1967 the Canadian chemical industry received over \$40 million from sales of pesticides (Friesen, 1969). Seventy four percent of this revenue represented sales of herbicides.

Huffaker (1964) listed some of the types of losses caused by weeds:

"1. Crowding out or reducing the growth of desirable plants, causing losses in yield and quality.

- 2. Much of the cost of cultivation.
- 3. The need for special seed and grain cleaning.
- 4. Direct injury to man, livestock, or livestock products.
- 5. Depreciation of watershed and wild-life values.
- 6. The serving as alternate hosts for insect pests or plant pathogens."

In contrast to the losses inflicted by weeds, there are occasions when the same species, in situations in which they are still considered to be weeds may be of some benefit to man. Van Den Bosch and Telford (1964) cited examples of the value of weeds in ensuring the continued success of biological control by entomophagous insects. For example, certain predatory insects pass a period of dormancy among the weeds along the edges of alfalfa fields and they are destroyed or disturbed if the weeds are removed. Bennett (1955) acknowledged the value of weeds in soil conservation:

"Even weeds reduce erosion, although not so effectively as grass. As compared with corn, weeds have caused a reduction of runoff on some types of land by more than a third and of erosion by more than a half, according to actual measurements. Russian thistle, at one time considered to be an insufferable pest in the Great Plains and other low-rainfall areas of the West, has proved itself a helpful plant for checking soil drift by wind. It is also of value as an emergency food in time of prolonged drought."

Both the costs and benefits that can be attributed to weeds are related to their life histories and to the type of environment provided by man in an agricultural situation. There are therefore many ways in which an appreciation of the ecology of a weedy species can be of value in determining the economics of its control.

The economics of weed control rest upon determining an

optimal balance between the costs of weed control and the benefits of a reduction in weed numbers (and thus a reduction in the losses that result from tolerating weeds). It is thus essential that some knowledge of the way in which a weed inflicts losses should be available. Competition between a crop species and a weed for water, light or mineral nutrients will be influenced greatly by the relative numbers of each species. The pattern that competition takes as the growing season progresses will depend upon the way in which the two species utilise the resources they obtain. The outcome may also depend upon which of the species had the earliest access to the contested resources.

The costs of controlling weeds may also have ecological implications. Species of weeds with underground rhizomes may have a greater resistance to mechanical cultivation than non-rhizomatous weeds. Weeds that are closely related, taxonomically, to the crop species may respond in the same way as the crop to the herbicides that are available. Weeds with seeds that germinate over a long period of time may require more frequent cultivation or application of herbicides for their control than weeds with seeds that germinate at one time only.

A knowledge of the ecological characteristics of a weed may suggest alternative methods of control that are less costly or provide greater benefits. For example, the discovery of a herbivorous insect that attacks a troublesome

weed may reduce the need to use a synthetic herbicide that has undesirable side-effects. The relationships between crop and weeds may be influenced by other variables in the environments they share. The costs and benefits of control may vary as such variables as climate, soil type, associated fauna, etc. influence the relative performances of crop and weeds.

In addition to a fuller understanding of the losses caused by weeds, research into the relationships between weeds and their environments can be expected to reveal any further beneficial effects that these species may have upon man.

The preceding discussion illustrates the value of ecological research as a basis for determining the most economic balance between the control and tolerance of weeds. This balance will shift towards increased control and less tolerance of weeds as the earth's finite resources are expected to meet the increasing per capita demands of a human population that at the present rate of increase will double in size in the next 35 to 37 years (Ehrlich and Ehrlich, 1970). With these prospects the need is even greater for a sound ecological basis on which to make economic decisions.

# Important aspects of the ecology of weeds

For a plant species to hehave as a weed of agriculture, it must possess adaptations that correspond to the characteristics of the agricultural environment. The

major aspects of the relationship between a weed and the agricultural environment can be listed:

- 1) The species must be introduced into the agricultural environment in sufficient numbers to cause economic losses.
- 2) Individual weeds must begin their growth at a time when competition with the crop species for such resources as light, moisture and mineral nutrients is minimal.
- The weed must be able to withstand competition from the crop as the latter increases its demands upon the available resources.
- 4) The weed must be able to withstand cultural techniques that are directed towards its elimination.
- 5) After the crop is harvested, the weed must be able to maintain its presence until favourable conditions recur.

#### The role of the seed

Several of the relationships listed above may involve the behaviour of the seeds. For a large number of annual weeds, seeds are the sole means whereby the species is introduced into a new environment and also whereby the species is maintained within the same environment from one season to the next. Control of the germination of the seeds may ensure that growth of the weed begins at the most favourable stage in the agricultural cycle. The existence of a reservoir of ungerminated seeds in the soil may enable the species to withstand cultivation and also may provide an

insurance against seasons in which the species is unable to produce new seeds successfully.

Although seeds enable the species to withstand unfavourable environmental conditions, the seeds themselves are influenced by environmental conditions. The way in which environmental variables influence the behaviour of seeds may have important implications for the success of the plants that subsequently develop from the seeds. Thus a study of the behaviour of the seeds of a weed may provide many insights into the characteristics of the species that are responsible for its success as a weed.

#### Objectives in this study

The present study was planned to achieve the following objectives:

- 1) To gain further insights into the ways in which weed species are adapted to the agricultural environment.
- 2) In particular, to determine the ways in which the behaviour of seeds contribute to the success of a species as a weed and to investigate the influence of environmental variables upon the behaviour of seeds.
- To relate such knowledge to the overall expression of success of a species, i.e. to its distribution as a weed of agriculture.

## The choice of species

The species that were chosen to be included in this investigation were <u>Amaranthus hybridus</u> L., <u>A. powellii</u>
S. Wats. and <u>A. retroflexus</u> L. The species were chosen

because each is an annual that is entirely dependent on seeds for reinfestation and colonisation and because together they are considered economically important weeds in Southwestern Ontario. The existence of three closely related and morphologically similar species provided an opportunity to compare the adaptations that permitted each species to behave as a weed. Small differences in the morphology of the three species may be reflected in subtle differences in behaviour and it was hoped that such differences might be more easily appreciated in a comparative study than in separate studies of each species.

Each of the species produces seeds over an extended period of time and thus seeds are exposed to much variation in environmental conditions both before and after maturing. It was felt that this characteristic made these suitable species in which to study the influence of environmental variables upon the behaviour of seeds.

There have been few studies in the past that dealt with the ecology of these three species. The germination behaviour of seeds of Amaranthus retroflexus has been the subject of several physiological investigations. However, these studies have been confined mostly to laboratory experiments and it is the exceptional author who has made attempts to relate his findings to the behaviour of the species in the field.

In contrast to the limited amount of information that is

A. hybridus and A. powellii. Indeed until recently (e.g. Sauer and Davidson 1961 and McWilliams 1966) there has been little appreciation of the widespread distribution and frequency of these two species as weeds in North America. This situation probably reflects both changes that have occurred in the distribution of the two species in the past century and the difficulties that have been encountered in identifying the three species.

#### Means of achieving the stated objectives

Two different approaches were followed in the present investigation. One approach was to describe the occurrence of the species in the field and to attempt to relate this to the range of variation among agricultural environments within the area studied. The second approach was to manipulate plants or environmental conditions or both in order to examine the effects of varying individual environmental variables or sets of variables. This approach was employed extensively in studies of the environmental control of seed behaviour. Subsequently attempts were made to integrate the information acquired from the different approaches.

The experimental work was planned also to investigate the variability of responses among different representative samples of plants of each species. To complement this aspect of the work, a taxonomic study of morphological variation within the three species was included.

#### CHAPTER 2

# THE TAXONOMY OF AMARANTHUS HYBRIDUS, A. POWELLII AND A. RETROFLEXUS

#### 2.1 Introduction

Amaranthus hybridus L., A. powellii S. Wats. and

A. retroflexus belong to the section Amaranthus (formerly

Amaranthotypus Dumort) which includes the type species

A. caudatus L. (Sauer, 1967). The characteristics of

this section have been described by Sauer (1967) as

follows:

"The section is distinguished from the bulk of the genus by the following combination of characters: plants monoecious; cymes continuing above uppermost leaves to form large, compound terminal inflorescences; tepals and stamens 5 (or varying between 3-5 in flowers of the same plant); utricle circumscissile (indehiscent in occasional mutant and hybrid individuals). As a rule these characters are extremely constant (the parenthetically noted variants are mainly within one species, A. powellii)."

The genus, and particularly this section, has often been considered taxonomically "difficult" (Grant, 1959, Brenan, 1961, Murray, 1940). Sauer (1967) pointed out that most of the difficulties have arisen from attempts to classify on the basis of growth form, which is under strong environmental control, and of characters that segregate within populations such as pigmentation. He

described reliable characters that in combination reveal genetically isolated species. He cautioned that hybridisation involving most members of the section has been reported and that some weedy and cultivated populations have patterns of variation which suggest introgression. Brenan (1961) expressed the following opinions:

"Many species of Amaranthus are remarkably similar to one another in general appearance. In the past this has led to the widespread use of certain familiar names for species to which they do not belong, and consequent confusion, both nomenclatural and taxonomic."

In the following pages a brief description will be given of each species. This will be accompanied by a discussion of synonyms that appear to describe the same species.

- 2.2 The individual species
- 2.2.1 Amaranthus hybridus L.

Sauer and Davidson (1961) gave the following description:

"Erect, coarse herbs, sometimes much branched. Leaves medium sized, generally at least 15 cm long when mature, ovate, rhombic oval, or lanceolate, usually acute. Flowers monoecious, in slender, lax, terminal, panicle-like thyrses, with many short, crowded lateral branches. Bracts slightly exceeding tepals and utricles, with medium thick, long-excurrent midribs. Tepals 5, those of pistillate flowers about equalling utricle, with simple midveins, acute, straight. Stamens 5. Style brances rather short, erect. Utricle rugose, circumscissile. Seed about 1 mm in diameter, dark brown, shiny."

Brenan distinguished three subspecies of A. hybridus:

(a) subsp. hybridus which includes the specimen in the

Linnaean Herbarium, No. 1117.19; (b) subsp. <u>incurvatus</u> which includes var. <u>incurvatus</u> and var. <u>cruentus</u>; and (c) subsp. <u>celosioides</u>. The distinction between the three subspecies was made on the basis of the relative length of the longer bracteoles of the female flowers to the length of the perianth:

- subsp. <u>hybridus</u> Longer bracteoles mostly twice as long as the perianth;
- subsp. <u>incurvatus</u> Longer bracteoles mostly as long as to 1½ times as long as the perianth;
- subsp. <u>celosioides</u> Longer bracteoles shorter than the perianth.

Sauer and Davidson (1961) commented on the relative length of tepals (= female perianth segments) to bracts (= bracteoles), but not to <u>longer</u> bracts. Their description is:

"bracts slightly exceeding tepals and utricles . . . "

From this description, it appears that they would definitely exclude subsp. celosioides from their concept of A. hybridus; they might exclude subsp. hybridus, but they would certainly include subsp. incurvatus. A further description of subsp. hybridus by Brenan (1961) suggests that Sauer and Davidson (1961) would exclude this subspecies from A. hybridus:

"In Britain A. hybridus var. hybridus is most likely to be confused with A. retroflexus from which it is usually readily distinguished by the sparser pubescence, the inflorescence running out into ± cylindrical tail-like spikes, and by the non-spatulate female perianth segments."

The reference to the inflorescence running out into tail-like spikes does not agree with Sauer and Davidson's description of panicle-like thyrses, with many short, crowded lateral branches. As will be seen later, Brenan's description coincides more closely with <u>A. powellii</u> as described by Sauer and Davidson (1961).

Aellen (1959) followed a different classification for A. hybridus. According to him, A. chlorostachys includes the synonym A. hybridus but not A. patulus. Brenan (1961) included A. patulus as a synonym for A. hybridus subsp. incurvatus. Aellen (1959) considered A. patulus as a separate species and his illustrations of the fruit of this species and of A. chlorostachys can be compared with Sauer's (1950) illustration for A. hybridus. Sauer's A. hybridus resembles A. patulus more closely than it does A. chlorostachys.

On the basis of the descriptions given, it appears that A. hybridus as conceived by Sauer and Davidson (1961) agrees most closely with subsp. incurvatus in Brenan (1961). It remains to evaluate the further subdivision of subsp. incurvatus into varieties. To quote Brenan:

"The var. cruentus (at least as far as Britain is concerned) comprises those variants with red or purple inflorescences. The structure of the inflorescences is, however, decidedly variable."

Sauer and Davidson (1961) treat <u>A. cruentus</u> L. as a separate species that differs from <u>A. hybridus</u>, <u>A. powellii</u> and <u>A. retroflexus</u> in the possession of bracts that are shorter than the utricle.

As will be seen in Chapter 4 (page 106) the majority of specimens of A. hybridus collected in Ontario have been assigned to subsp. incurvatus (Frankton, personal communication). Frankton has determined three specimens from the province as subsp. hybridus and these have been confirmed by Sauer as A. hybridus but without recourse to further sub-division. Frankton had this to say of the three specimens:

"[compared with specimens of subsp. incurvatus, they] had longer bracteoles, a more bristly appearance in the inflorescence, fewer branches and greener foliage and furthermore had teardrop-shaped seeds with length/width ratios of about 5/4. Subsp. incurvatus with shorter bracteoles had seeds almost perfectly round."

The use of the name A. hybridus throughout the remaining chapters of this thesis will be taken to mean specifically subsp. incurvatus var. incurvatus unless otherwise stated. Plants of this species that were included in the present investigations, and specimens observed in the field agreed with the description of A. hybridus by Sauer and Davidson (1961) and the description of subsp. incurvatus var. incurvatus by Brenan (1961).

The following synonyms were listed by Brenan (1961) for A. hybridus L. subsp. incurvatus (Timeroy ex Gren. & Godr.):
A. hybridus subsp. incurvatus:

A. cruentus L.

A. hybridus L. subsp. cruentus (L.) Thell.

A. hybridus subsp. incurvatus var. incurvatus:

A. patulus Bertol.

A. incurvatus Timeroy ex Gren. & Godr.

A. patulus Bertol. subsp. <u>incurvatus</u> (Timeroy ex Gren. & Godr.) Arc.

A. hybridus L. subsp. cruentus (L.) Thell. var. patulus (Bertol.) Thell.

### 2.2.2 Amaranthus powellii S. Wats

Sauer and Davidson (1961) provided the following description of this species:

"Erect, coarse, much branched herbs. Leaves rather small, generally under 10 cm long, ovate, rhombic-oval, or lanceolate, usually obtuse. Flowers monoecious, in long, thick, stiff, spike-like terminal thyrses, some with a few, long, widely-spaced lateral branches. Bracts far exceeding tepals and utricles, with thick, moderately excurrent midribs. Tepals 3-5, the longer ones of the pistillate flowers exceeding the utricle, with simple midveins, acute, nearly straight. Stamens 3-5. Style branches long, recurved from base. Utricle slightly rugose, circumscissile. Seedabout 1¼ mm in diameter, dark brown, shiny."

Sauer (1967) stated that A. powellii is an abundant weed in Europe, where it is usually misidentified as

A. chlorostachys Willd. which is a synonym of A. hybridus.

He gave as a synonym, A. chlorostachys Willd. var.

pseudoretroflexus Thell. Brenan (1961) made no mention of

A. powellii but he did include A. chlorostachys var.

pseudo-retroflexus as a synonym for A. hybridus subsp.

hybridus var. pseudo-retroflexus, with this description:

"has large bracteoles usually 5mm or more long, and stout dense inflorescences giving the plant an aspect recalling that of A. retroflexus (typical var. hybridus having smaller bracteoles about 3-5mm long and more slender inflorescences)."

Sauer (1967) used the absolute length of the bract to distinguish between A. powellii and A. hybridus, quoting 5 mm for the former and 3-4 mm for the latter. These figures correspond respectively with those given by Brenan (1961) for A. hybridus subsp. hybridus var. pseudo-retroflexus and typical subsp. hybridus. There is close agreement between the two sets of figures.

Brenan (1961), in his discription of <u>A. hybridus</u> subsp. <u>hybridus</u>, mentioned a "second minor variant", var. <u>pseudo-retroflexus</u> (Thell.) Thell. subvar. <u>aristulatus</u> Thell. which he characterised in the following way:

"having most of the female perianth-segments bearing at their apex "a distinct awn-like point about ½ mm long"."

He continued:

"Neither of these variants, however, seems to me of much taxonomic significance, or to be more than a mere form of var. hybridus."

He concluded his discussion of subsp. <a href="https://hybridus">hybridus</a> by reporting that the specimen of <a href="https://hybridus">A. hybridus</a> in the Linnaean Herbarium, No. 1117.19, which is typical subsp. <a href="https://hybridus.hybridus">hybridus</a>, is not var. <a href="https://pseudo-retroflexus">pseudo-retroflexus</a> nor subvar. <a href="https://aristulatus.sauer">aristulatus</a>. Sauer (1967) stated that specimen 1117.19 agrees with other material available for typification of <a href="https://www.hybridus.hybridus">A. hybridus</a>. However, the description of <a href="https://www.hybridus.hybridus">A. hybridus</a> subsp. <a href="https://hybridus.hybridus.hybridus">hybridus</a> given by Brenan (1961) resembles closely Sauer and Davidson's (1961) description of <a href="https://www.hybridus.hybridus.hybridus.hybridus.hybridus">hybridus</a> subsp. <a href="https://www.hybridus.

cylindrical tail-like spikes..." (Brenan) and "long, stiff, spike-like terminal thyrses, some with a few, long, widely spaced lateral branches." (Sauer and Davidson).

Aellen (1959) considered that A. chlorostachys consists of six varieties of which two are: var. pseudoretroflexus (for which the synonym A. powellii is given), and var. aristulatus. His illustration of the fruit of A. chlorostachys (Aellen, 1959, Fig 205) does not agree entirely with Sauer's (1950) illustration for A. powellii, but the similarity is greater than with Sauer's illustration of A. hybridus. However, the illustration of A. chlorostachys does represent better some of the collections of A. powellii included in the present investigation than do Sauer's illustrations.

The material of A. powellii used in this investigation exhibited a considerable range of variation which is discussed more fully in Chapter 3. Although this variation was discontinuous, it fell within the range described for this species by Sauer and Davidson (1961), for this reason all the material will be described as A. powellii. To distinguish between variants, use will be made of the term 'type', and within 'types' and within species the term 'collection' will be used to describe seed collected from different localities. It is possible that some of the variation observed in local collections of A. powellii can be explained in terms of the distinction made between A. chlorostachys var. pseudoretroflexus and var. aristulatus by Aellen (1959).

The following is a summary of what appear to be synonyms of <u>A. powellii</u> (Aellen, 1959, Brenan, 1961, Sauer, 1967):

Amaranthus chlorostachys Willd. var.

pseudoretroflexus (Thell.) Aellen

Amaranthus chlorostachys Willd. var. (ssp.)

powellii Priszter

Amaranthus hybridus L. subsp. hypochondriacus

(L) Thell. var. chlorostachys (Willd.) Thell.

for. pseudoretroflexus Thell.

Amaranthus hybridus L. subsp. hybridus var.

pseudo-retroflexus (Thell.) Thell.

Amaranthus hybridus L. subsp. hybridus var.

pseudo-retroflexus (Thell.) Thell. sub var.

aristulatus (?)

Amaranthus chlorostachys var. aristulatus (Thell.)

Aellen (?)

Amaranthus bouchoni Thell. (See Sauer, 1967).

2.2.3 Amaranthus Retroflexus L.

Sauer and Davidson (1961) gave the following description of the species:

"Erect, coarse herbs, sometimes much branched.
Leaves medium-sized, generally at least 15 cm long,
when mature, ovate, rhombic-oval, for lanceolate,
usually obtuse. Flowers monoecious, in thick, stiff,
panicle-like terminal thyrses, with many short,
crowded lateral branches. Bracts far exceeding
tepals and utricles, with thick, shortly-excurrent
midribs. Tepals 5, those of pistillate flowers
exceeding the utricle, with simple mid-veins, obtuse
or emarginate, recurved. Utricle slightly rugose,
circumscissile. Seed 1 mm in diameter, dark brown,
shiny."

This species has escaped the nomenclatural confusion which taxonomists have bestowed upon the two preceding species. Brenan (1961) described two varieties, var. retroflexus and var. delilei which are distinguished on the length of the bracts (shorter in delilei). However, Brenan pointed out that the two are connected by intermediates and the value of the distinction is thus questionable.

Neither Aellen (1959) nor Brenan (1961) nor Sauer. (1967) gave synonyms for this species.

### 2.3 Hybridisation

To complicate further the descriptions of the species and the identification of unknown material, there are many reports of natural and artificial hybrids between these three species and with other species of Amaranthus (Murray, 1940, Sauer, 1957, 1967, Tucker and Sauer, 1958, Priszter, 1958, Sauer and Davidson, 1961, McWilliams, 1966, McWilliams, Landers and Mahlstede, 1968). This discussion will be restricted to hybridisation between the three species under investigation and will be followed by a list of reports of hybridisation with other species.

Murray (1940 and personal communication) demonstrated hybridisation experimentally as part of an investigation into the determination of sex in dioecious species of the genus. He reported that the monoecious species he was using (including A.hybridus, A. powellii and A. retroflexus)

readily hybridised but produced highly sterile F1 plants that made it virtually impossible to obtain F2 plants. All species of the genus are wind pollinated and he estimated that self-pollination takes place in 85% and cross-pollination in 15% of successful fertilisations. In any plant the stigmas of the first female flowers are receptive before any male flower has opened thus providing the potential for cross-pollination and hybridisation. With an isolated plant of one species surrounded by many plants of a different species it was possible to measure the interspecific hybridisation between A. hybridus and A. retroflexus.

Tucker and Sauer (1958) described some Californian populations of Amaranthus whose origin they attributed to recent hybridisation between the three species under consideration and two further species, A. caudatus and They reported the occasional occurrence of A. cruentus. highly sterile plants that were morphologically similar to Murray's synthetic hybrids. They observed other plants that were also apparent hybrids and some of these showed Many of these individuals revealed no sterility at all. combinations of characters from three of the five species. Characters of A. powellii and A. retroflexus occurred in all of the hybrids showing three-species combinations, together with characters of one of the other species. One collection of 21 specimens included a variety of individuals with many combinations of characters of

A. hybridus, A. powellii and A. retroflexus. Three of these plants were sterile and the remainder were noteworthy in possessing utricles that were not circumscissile; a character belonging to none of the five species involved in the complex.

Sauer and Davidson (1961) commented on the occurrence of hybrids in Wisconsin:

"Recently A. powellii has begun mixing with A. retroflexus, and some sterile hybrids have resulted. All of these were collected since 1922 in artificially disturbed sites."

Priszter (1958) gave European records for hybrids of the following combinations of species: A. chlorostachys var. genuinus x retroflexus; A. chlorostachys var. powellii x retroflexus; and A. patulus x retroflexus.

Sauer (1967), discussing the geography of  $\underline{A}$ , powellii, noted:

"The species is involved in partially fertile hybrid swarms in eastern North America, where it is invading territory of A. retroflexus and A. hybridus, in western North America where the converse is true, and in Europe, where all three are recent immigrants."

McWilliams, Landers and Mahlstede (1968) reported further evidence of hybridisation in studies of seed weight and dormancy. The seeds of <u>A. powellii</u> are heavier and more dormant than those of <u>A. retroflexus</u>. Putative hybrids were found which were intermediate in morphology, seed weight and dormancy. The authors suggested that hybridisation with <u>A. powellii</u> may be responsible for the occurrence of populations of <u>A. retroflexus</u> with seed dormancy.

### Hybridisation with other species

The following table (Table 2.1) lists records of hybrids between the three species; A.hybridus, A. powellii and A. retroflexus and other species of the genus.

### TABLE 2.1

A LIST OF THE HYBRIDS INVOLVING A. HYBRIDUS,

A. POWELLII OR A. RETROFLEXUS THAT HAVE BEEN REPORTED IN THE LITERATURE.

### SPECIES INVOLVED

### REPORTED IN

### A. hybridus

A. hybridus L. x quitensis H.B.K.

Murray, 1940<sup>xa</sup>

A. hybridus L. x tamariscinus Nutt.

Murray, 1940<sup>xb</sup>, Sauer, 1957

A. hybridus L. x tuberculatus Moq.

Sauer, 1957

### A. powellii

A. australis A.Gray x powellii S.Wats. Murray, 1940 xb

A. caudatus L. x powellii S. Wats.

Tucker and Sauer, 1958

A. leucocarpus L. x powellii S. Wats.

Murray, 1940<sup>xa</sup>

A. powellii S.Wats x quitensis H.B.K.

Murray, 1940<sup>xa</sup>

A. powellii S.Wats. x tamariscinus Nutt.Murray, 1940 xb

A. powellii S.Wats. x tuberculatus Moq. Murray, 1940 xb Sauer and

Davidson, 1961

### A. retroflexus

A. australis A. Gray x retroflexus L.

Murray, 1940<sup>xb</sup>

### SPECIES INVOLVED

A. caudatus L. x retroflexus L.

Tucker and Sauer, 1958

REPORTED IN

A. caudatus L. x powellii S.Wats x retroflexus L.

Tucker and Sauer, 1958

A. cruentus L. x retroflexus L.

Murray, 1940<sup>xa</sup> Tucker and Sauer, 1958

A. cruentus L. x powellii S.Wats x retroflexus L.

Tucker and Sauer, 1958

A. hypochondriacus L. x retroflexus L.

Priszter, 1958

A. paniculatus L. x retroflexus L.

Priszter, 1958

A. quitensis H.B.K. x retroflexus L.

Murray, 1940<sup>xa</sup>

A. retroflexus L. x spinosus L.

Murray, 1940

A. retroflexus L. x tamariscinus Nutt.

Murray, 1940<sup>xb</sup>

A. retroflexus L. x tuberculatus Moq.

Murray, 1940<sup>xb</sup> Sauer, 1957, Sauer and Davidson, 1961

Notes: xa - see Sauer (1953) for revised identification of Murray's material.

xb - see Sauer (1955) for inclusion of genus Acnida in Amaranthus

Only one of the above mentioned species, <u>A. tuberculatus</u>, occurs naturally within the area from which material for this investigation was collected. Concerning hybridisation of this species with <u>A. hybridus</u> and <u>A. retroflexus</u>
Sauer (1957) has written:

"there is no indication that these hybrids have bred successfully, either among themselves or with the parent species. Murray reported that over 90% of their pollen was visibly defective under the microscope, as compared to somewhat over 50% in monoecious x monoecious or dioecious x dioecious interspecific hybrids."

A. tuberculatus is a dioecious species, as are
A. australis and A. tamariscinus.

Of the other species that enter into hybridisation,

A. caudatus, A. cruentus and A. hypochondriacus could

conceivably be of local importance. Sauer and Davidson

(1961) have the following to report of their occurrence

in Wisconsin:

"These species are now grown as ornamentals around the world and often sold in commercial flower seed packets. All three have undoubtedly been repeatedly planted and occasionally escaped in Wisconsin, although rarely collected."

From the descriptions of hybrids involving these species given by Tucker and Sauer (1958), it would appear that any hybrids with such an origin would be readily identified by their intense pigmentation.

Before leaving the subject of hybridisation, it is worth noting some comments made by Sauer (1957) on the identity of hybrids between A. tamariscinus and

### A. tuberculatus:

"Beyond establishing the fact of hybridisation lies the greater problem of tracing its consequences. These are not fully assessible because it takes only a few back-crossings before the descendants of an obvious hybrid begin to look like pure examples of the recurrent parent."

### CHAPTER 3

### A QUANTITATIVE DESCRIPTION OF THE THREE SPECIES IN SOUTHWESTERN ONTARIO

### 3.1 Introduction

There were three separate reasons for making a taxonomic survey of the species as they occur in South-western Ontario.

Firstly there was a need to describe precisely the morphological differences between collections of A. powellii and A. retroflexus used in other experiments. When plants were grown under uniform conditions at the Department of Botany Experimental Farm there was much variation in morphology between the individuals of different collections of nominally the same species. Much of the variation was in growth form but it also included variation in some of the characters that have been considered diagnostic for the species.

The second reason arose from the results of germination experiments (particularly expts. 9 and 10 described in Chapter 7) in which it was demonstrated that collections of A. powellii could be placed into at least two groups on the basis of their germination response. Collections

that were similar in response came from widely separated localities. It was thus desirable to discover whether dissimilarities in germination response reflected morphological differences and to determine more precisely their geographical status.

Thirdly, there was a need to determine whether evidence existed of hybridisation and introgression between the species in Southwestern Ontario.

### 3.2 Materials

Preserved specimens were available from several different sources which are listed in the following table (Table 3.1).

TABLE 3.1

THE SOURCES OF SPECIMENS EXAMINED

IN THE TAXONOMIC INVESTIGATION

TIME OF COLLECTION	SOURCES OF MATERIAL
Fall 1966	Specimens from the field accompanying collections of seed used in germination studies (see chapters 6 and 7).
Summer 1967	Plants grown under uniform field conditions from seed collected in the autumn of 1966.
Summer 1967	Specimens collected during the survey of the distribution of the species (see chapter 5).
Summer 1968	Plants grown under greenhouse conditions from seed collected during the 1967 survey of distribution.
Fall 1968	Specimens collected from the field to supplement existing material.

Figure 3.1 indicates the localities from which specimens and seeds were collected. It will be seen that collections of A. powellii have been assigned to one of two 'types'. This was done on the basis of the morphology of the fruit, by visual inspection. This distinction was made before analysis to allow these individuals to be identified more easily throughout the stages of analysis. It also provided a test of the accuracy with which specimens could be distinguished without recourse to precise measurement.

It was necessary to strike a balance between describing a large number of specimens from one site and sampling a representative number of sites. Usually three plants were sampled from each site. When there appeared to be variation between the plants of one species at one site, a larger number of plants was sampled.

It is noted in table 3.1, that not all material was collected directly from the field. Some collections were represented by plants grown under uniform field conditions and others were represented by plants grown in the greenhouse. In a few collections, specimens were available from both the field and artificial culture.

A list of the specimens examined is presented in table 3.2.

Information is included describing the site and briefly the habitat of each collection.

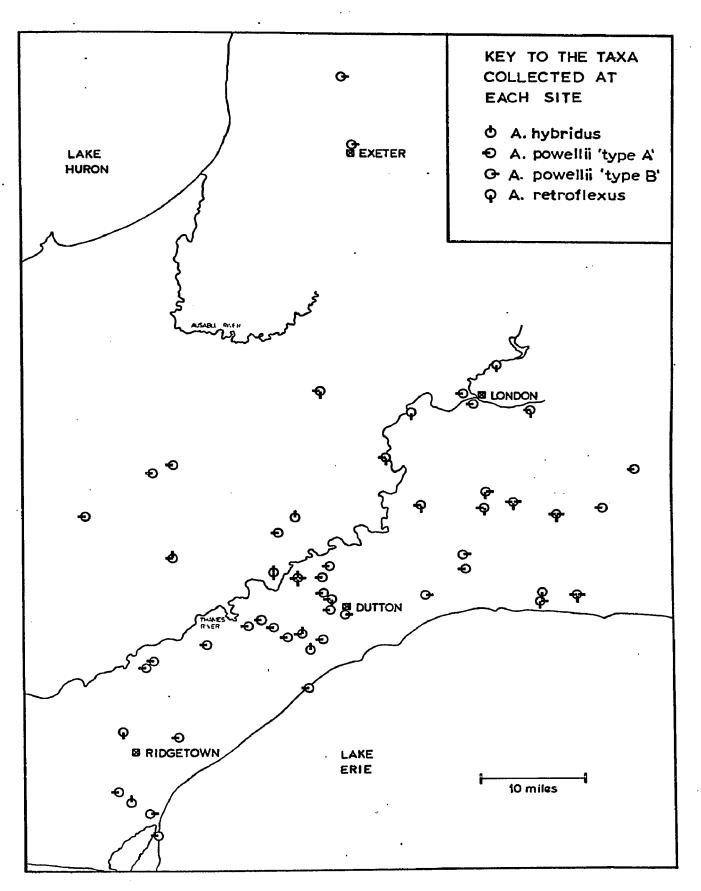


Fig. 3.1 A map of the sites from which specimens examined in the taxonomic survey (or seeds from which they were grown) were collected.

Species and Culture\*

### TABLE 3.2

A LIST OF THE SPECIMENS INCLUDED IN THE TAXONOMIC INVESTIGATION TOGETHER WITH DETAILS OF THE ORIGINAL HABITAT FROM WHICH THE SPECIMENS (OR THE SEEDS FROM WHICH THEY WERE GROWN) WERE COLLECTED

### Details of original habitat

County

Crop Soil\* Grid\*

sample no.	Ouroure	2 OTOD	POTT	reference	County
A. hybridus 1-3 4 5 6 7-9 10-12 13,14	B.E.F. FIELD FIELD G.H. G.H. G.H.	Soybeans Soybeans Buckwheat Soybeans Tobacco Soybeans Soybeans	BSL HC BEL BEL BEL BCL BSL	264915 518390 522288 483297 541169 379323 528195	Kent Middlesex Elgin Middlesex Elgin Middlesex Elgin
A. retroflexus 15-17 18-20 21-23 24 25 26,27 28-30 31-33 34-36 37-39 40,41 42-44 45-47 48-50 51-53 54-56 57-59 61-63 64-65	B.E.F. B.E.F. B.E.F. B.E.F. FIELD	Fallow (Roadside) Potatoes Soybeans Buckwheat Soybeans Tobacco Corn Soybeans Soybeans Potatoes Soybeans Corn Soybeans Soybeans Corn Soybeans Corn Soybeans Corn Soybeans Corn Soybeans Corn Coybeans Corn Coybeans Corn Coybeans Corn Coybeans Corn	BFFHBCBTMMBGBFLMMHC BFFHBCBTMMBGBFLMMHC	252035 697566 828645 518390 522253 572258 573258 556602 043394 896248 898263 71440 657490 8094432 853445 880569 445208	Kent Middlesex Middlesex Middlesex Elgin Elgin Elgin Middlesex Elgin Elgin Elgin Elgin Elgin Elgin Elgin Middlesex Middlesex Elgin Middlesex Elgin Middlesex Elgin Middlesex Elgin Middlesex Elgin
A. powellii 'type A' 66 67 68 69 70 71 72-74 75-77	FIELD FIELD FIELD FIELD FIELD FIELD FIELD FIELD FIELD	(Riverbank) Soybeans (Roadside) (Landfill) Corn Buckwheat Tobacco Corn	BCL BES FG HC BEL BCL TS	792580 244934 304860 777598 493365 522288 953258 556602	Middlesex Kent Kent Middlesex Middlesex Elgin Elgin Middlesex

(continued)

### Details of original habitat

Species and sample no.	Culture* Crop	Soil* Grid* reference	County
78-80 81-88 84-88 89,93 94-96 97-96 97-98 91-97 105-10	FIELD Soybeans FIELD Soybeans FIELD Soybeans FIELD Soybeans FIELD Soybeans FIELD Soybeans FIELD Corn FIELD Soybeans G.H. Corn G.H. Corn G.H. Soybeans	LSL 300476	Elgin Elgin Elgin Middlesex Middlesex Elgin Elgin Middlesex Elgin
A. powellii 'type B' 177-180 181-183 184 185 186-188 189-191 192-194 195-197 198	B.E.F. Turnips B.E.F. Soybeans FIELD FIELD Buckwhea FIELD Tobacco FIELD Soybeans FIELD Potatoes FIELD Soybeans FIELD Soybeans	As samples 177-180 t BEL 522288 BCL 953258 MC 923394 BSL 896248 ML 812432	Huron Huron Elgin Elgin Elgin Elgin Middlesex Elgin

(continued)

### Details of original habitat

Species and sample no.	Cultur	e* Crop	Soil*	* Grid* reference	County
199,200 201-203 204-206 207-209	G.H. G.H. G.H.	Soybeans Corn Corn Corn	BVS BEL HL BVS	717259 592228 293895 775327	Elgin Elgin Kent Elgin
Putative hybrids 210 211 212	B.E.F.	These three appeared sp Amaranthus	ontane	eously in	plots of

### \*Key to abbreviations

### 1. Cultural conditions:

- B.E.F. Plants grown under uniform conditions at the Department of Botany Experimental Farm, as described on page 362.
- FIELD Specimens collected from various natural sites in Southwestern Ontario.
- G.H. Plants grown in potting soil in 25 cm (diameter) pots in greenhouse 19 under conditions described in table 6.3.

### 2. Soil types:

The soil type at the site of origin was determined from Soil Survey Report maps (Canada Department of Agriculture). The abbreviations used are the same as those given in Appendix 2, table A2.3.

### 3. Grid reference:

Reference figures give the position to the nearest 100 metres on the Universal Transverse Mercator Grid. The grid references in this table are between 4200 and 5050 East and between 46860 and 47650 North with the exception of samples 177 to 184 which are at either 48014 or 48128 North.

### 3.3 Characters Measured

The choice of characters was influenced by those used in previous taxonomic surveys of the genus (Tucker and Sauer, 1958, Sauer and Davidson, 1961, Brenan 1961, Sauer 1967) and also by the experience gained through cultivating the plants and handling the fruits in germination experiments. Table 3.3 is a composite of the characters used by earlier authors and the generalisations that they have made for each species in terms of these characters. Most of the characters have been expressed in qualitative terms. In some cases it would be easy to measure the same character in quantitative terms with a gain in information. In other cases the characters could only be quantified with difficulty.

The characters chosen for inclusion in this survey are presented in table 3.4. Characters were chosen that could be measured with speed and accuracy in all specimens. Characters that could not be expressed easily in quantitative form were avoided in order to simplify the preparation of data for analysis. The characters included describe the inflorescence, the bracts, the female flower and the fruit. The parts of the female flower persist with the fruit and it is possible to measure characters of flower and fruit at one time, from one fruit. Murray (1940) and Brenan (1961) have described the structure of the flowers and their arrangement on the inflorescence.

CHARACTERS THAT HAVE BEEN EMPLOYED BY

## EARLIER AUTHORS TO DIFFERENTIATE BETWEEN

# A. HYBRIDUS, A. POWELLII AND A. RETROFLEXUS.

STATE OF THE CHARACTER IN EACH SPECIES

AUTHOR \* CHARACTER

		A. HYBRIDUS	A. POWELLII	A. RETROFLEXUS
^	Terminal Inflorescence:			
1,2,4	Rigidity	lax	stiff	stiff
ζ-	length of terminal spike	short	long	short
<del>_</del>	Shape of terminal spike	moderately slend <b>e</b> r	thick	thick
1,2,4	Number of branches	numerous	few	numerous
1,2,4	Density of branches	crowded	widely-spaced	crowded
4	Shape of branches	slender	slender	thick

(continued)

STATE OF THE CHARACTER IN EACH SPECIES

AUTHOR\* CHARACTER

A. RETROFLEXUS		extremely long		1	twice as long		extremely thick	barely excurrent		very long	narrowly obovate, spathulate	recurved
A. POWELLII		extremely long	5 mm	twice as long	twice as long	equalling	very thick	excurrent		very long	oblong, lanceolate to narrowly ovate	straight
A. HYBRIDUS		moderately long	3-4 mm	equalling or a little longer	1-1½ times as long	shorter	medium thick	long-excurrent		medium	oblong, lanceolate to narrowly ovate	straight
	Bracts:	Length .	Length	Length relative to utricle	Length of longer bracts relative to perianth	Length of lamina relative to utricle	Thickness of midrib	Excurrence of the midrib	Female Perianth Segments:	Length	Shape	Habit
		7,2	7	īζ	23	CJ	1,2,4	7,2		~	۲. در	7,0

(continued)

STATE OF THE CHARACTER IN EACH SPECIES	A. HYBRIDUS A. POWELLII A. RETROFLEXUS	acute acute emarginate or obtuse to truncate	5 5-5 5	e shorter than to conspicuously slightly exceeding exceeding		erect recurved erect	slender stout moderately stout	r' moderately narrow broad no tower
	A. HYBRID	acute	7					
CHARACTER		Apex	Number	Length relative to fruit	Fruit:	Habit of style branches	Bases of style branches	Shape of 'tower'
AUTHOR*		1,2,3,	2,4	īζ		1,2,3	7,4	<b>~</b>

### \*Key to authorities:

 $\overline{\phantom{a}}$ 

1961.
3 - Brenan,
1961.
Sauer and Davidson,
2 <b>-</b> Sa
1958.
und Sauer,
- Tucker a

5 - Gleason and Cronquist, 1963.

4 - Sauer, 1967.

TABLE 3.4
A DESCRIPTION OF THE CHARACTERS MEASURED FOR

MAXIMUM VALUES RECORDED AMONG THE SPECIMENS.

EACH SPECIMEN TOGETHER WITH THE MINIMUM AND

CODE NO.	CHARACTER	MINIMUM VALUE	MAXIMUM VALUE
	<u>Utricle</u> :		
1	Distance from base to point of circumscission ("cup")	8.63 ua.*	18.25 ua.
2	Distance from point of circumscission to stylar sinus (cap)	6.88 ua.	13.00 ua.
3	Number of style branches	2.38	3.25
	Female Perianth:		
4	Number of tepals	3.50 ua.	5.25 ua.
5	Proportion of tepals with excurrent midrib	0.09	1.00
6	Proportion of tepals with retuse apex	0.00	0.95
7	Length of longest tepal	19.38 ua.	48.38 ua.
	Seed:		
8	Length	30.25 ub.	44.00 ub.
9	Relative distance from micropyle to point of greatest width	0.47	0.82
10	Greatest width	26.88 ub.	35.00 ub.
	Ratios:		
11	Utricle cup/utricle cap (position of circumscissi	0.86 on)	2.07
12	Seed width/seed length	0.75	0.99

CODE NO.	CHARACTER	MINIMUM VALUE	MAXIMUM VALUE
	Within Plant Variance:		
13	Variance of character 1	0.00 ua <sup>2</sup> .	4.57 ua <sup>2</sup> .
14	Variance of character 2	0.13 ua <sup>2</sup> .	2.57 ua <sup>2</sup> .
15	Variance of character 3	0.00	0.57
16	Variance of character 4	0.00 ua <sup>2</sup> .	0.98 ua <sup>2</sup> .
17	Variance of character 5	0.00	0.10
18	Variance of character 6	0.00	0.13
19	Variance of character 7	1.13 ua <sup>2</sup> .	76.13 ua <sup>2</sup> .
20	Variance of character 8	0.13 ub <sup>2</sup> .	15.64 ub <sup>2</sup> .
21	Variance of character 9	0.00	0.02
22	Variance of character 10	0.00 ub <sup>2</sup> .	15.13 ub <sup>2</sup> .
23	Variance of character 11	0.00	0.20
24	Variance of character 12	0.00	0.04
	Bract:		
25	Length	25.13 ua.	72.13 ua.
26	Variance of character 25	1.55 ua <sup>2</sup> .	413.0 ua <sup>2</sup> .
27	Utricle cup+cap/bract length	0.36	0.83
28	Length of longest tepal/ bract length	0.45	1.12
	Inflorescence:		
29	Length of terminal spike	9 mm	307 mm
30	Proportion of terminal spike in which stem is visible	0.00	0.88
31	Width of terminal spike 10 mm from apex	5 mm	32 mm
32 32	Width of terminal spike at mid length	5 mm	81 mm

CODE NO.	CHARACTER	MINIMUM VALUE	MAXIMUM VALUE
33	Length of 1st lateral below terminal spike	6 mm	221 mm
34	Width of first lateral at mid length	4 mm	22 mm
35	Length of third lateral	7 mm	270 mm
36	Width of third lateral at mid length	4 mm	23 mm
37	Length of fifth lateral	6 mm	375 mm
38	Width of fifth lateral at mid length	3 mm	24 mm
39	Relative length of first lateral to the mean of three	0.13	1.35
40	Relative length of fifth lateral to the mean of three	0.53	2.34
41	Number of laterals in 100 mm below terminal spike	1	55
* <u>Units</u>	mm - millimetres		
	ua micrometer units objective (12 ua		<i>i</i> -power
	ub micrometer units objective (29 ub	with the high. = 1 mm)	gh-power

The characters numbered 1 to 12 are the means of observations made on eight fruits for each plant.

Wherever possible, the eight fruits were removed from the

variance units  $(144 \text{ ua}^2 = 1 \text{ mm}^2)$ 

variance units (841  $ub^2 = 1 mm^2$ )

ua<sup>2</sup>. -

ub<sup>2</sup>. -

terminal spike of each inflorescence without the aid of a microscope. Each fruit was examined under a binocular dissecting microscope and measurements were made using a micrometer eye-piece.

The mean of eight observations of a character was more representative of the state of that character in the plant than a single observation. However some information concerning the variability of a character was lost when the eight observations were replaced by their mean. Consequently it was decided to include the variance of each character as an additional character. Characters 13 to 24 are the variances of characters 1 to 12 respectively.

Character 25, the length of the bract, is also the mean of eight observations for each plant. Character 26 is the variance of character 25. Characters 27 and 28 are ratios of the mean values for each of the characters they include.

Characters 29 to 41 describe the morphology of the terminal inflorescence and thus are based on one observation for each plant. These characters were measured without the aid of a microscope; measurements were taken to the nearest millimetre.

### 3.4 Methods of Analysis

The purpose of this investigation was not to produce a classification of the three species (which would be

meaningless in isolation from other species in the section) but rather to describe the nature of variation within and between them. Factor analysis is a general technique ideally suited to this need and the usefulness of a particular method, "Principal Components Analysis", has been stated by Ivimey-Cook (1969):

"This technique investigates the relationships between a multidimensional array of variables, some of which may be correlated and hence at least partially redundant. The points representing the variables form a roughly hyperellipsoidal swarm and the method extracts a set of orthogonal, and hence uncorrelated, components, these being the principal axes of the hyperellipsoid. The components are extracted in descending order of magnitude and much of the variance is accounted for by the first few components, in effect reducing the dimensions of the problem."

The principal components are extracted from a correlation matrix (or other type of similarity matrix) The choice of matrix constructed from the original data. may be of importance. In the present investigation a matrix of correlation coefficients was favoured for the following reasons. One of the properties of the correlation coefficient (Snedecor and Cochran, 1968 section 7.2) is that it is a pure number without units or Since it is unlikely that the units of demensions. measurement contribute meaningful information to the analysis, choice of the correlation matrix will eliminate any effect that these units may have (Pearce, 1965). can be seen in table 3.4 that the characters included in this investigation were measured in several different units. The choice of the correlation coefficient also ensures that each variate is given equal weight when determining the amount of variation explained by a particular vector. The consequences of departures from this situation are described by Pearce (1965).

Two strategies are available for the execution of principal components analysis. The R-type analysis extracts components from a matrix of correlations between attributes (characters), whereas the Q-type analysis utilises a matrix of similarities between individuals (samples). Both analyses can lead to the same set of results (Orloci, 1966), and the R-type analysis is to be preferred when there are more samples than characters. This was the case in the present investigation with 211 samples described by 41 characters. A further advantage of the R-type analysis is that it allows additional samples to be added to an already completed ordination (Orloci, 1966).

A programme that executes an R-type factor analysis was available from the Computing Centre of the University of Western Ontario for use with their PDP 10/50 time-sharing system. This programme accepts raw data and computes the correlation matrix before executing the component analysis. Optional outputs include means and standard deviations, the correlation matrix, eigenvalues, the factor matrix and the factor scores.

An eigenvalue is computed for each of the component axes as these are extracted. This parameter is a measure of the variance extracted by each component (Jancey, 1966) and since components are extracted in order of the amount of variation they describe, the eigenvalues will decline for successive components extracted. The more rapidly the eigenvalues decline, the greater is the proportion of variation accounted for by the components first extracted. The proportion of variation accounted for by the first component reflects the degree of correlation between characters with the most variation.

It is of interest to determine which of the original characters have contributed the most variation to each of the principal components. This information is provided in the factor matrix, which consists of an expansion of the eigenvalue of each component into factor loadings. The factor loadings are measures of the amount of variation contributed by each character to each component. The positive or negative nature of a factor loading relates the direction of variation on the component axis to the direction of variation on the original character axis.

The factor scores enable each of the original samples to be ordinated on each new component axis. Since by definition the components are orthogonal it is possible to ordinate the samples on three axes at the same time and represent the results in three-dimensional drawings.

When the samples have been ordinated on a particular axis, it is possible to relate their position on the new axis to the values of the original characters in which they were measured. This is done by considering the factor loadings on the component axis in question. For example, samples that have a large positive score on the first component axis will have high measured values in those characters that contributed large positive factor loadings to the first axis. These samples will also have low measured values in those characters that contributed large negative loadings to this axis.

There are as many component axes that can be extracted as characters in the original data matrix. Since the amount of variation described by each component declines rapidly with successive components extracted, there is little value in considering more that the first This is indeed the purpose and value of component analysis, i.e. to reduce the number of dimensions in which the problem is described. There remains the question of deciding exactly how many components need to be No hard and fast rule can be given. one thing, the number that can usefully be considered will be inversely related to the rate at which successive eigenvalues diminish in value. It is often sufficient to consider the relationships between samples on the first three axes and this allows relatively easy representation

and interpretation. At least two recent authors have adopted the convention of examining in any detail only those components whose eigenvalues are greater than unity (Jancey, 1966, Ivimey-Cook, 1969).

When a decision has been made concerning the number of components that will be examined, it is then only necessary to consider factor loadings and factor scores for that number of axes. The programmed analysis that was used has facilities for determining the number of components that are to be extracted.

The purpose of the principal components analysis, as stated earlier is to define variation within the data in the most efficient terms. This it does well, but it does not "prove" anything nor attach significance to any differences. Pearce (1965) considers the technique to be a valuable tool for defining hypotheses. He cautions:

"The hypotheses evolved by multivariate methods are like any other and should be accepted only if they can be coordinated with other knowledge and can be confirmed by experimental evidence."

The results of ordinations are often employed to demonstrate discontinuities in variation within data and to suggest discrete taxonomic (or ecological) entities.

Although such conclusions may be valid, they are not a consequence of the analysis but rather of its interpretation. The value of principal components analysis in demonstrating discontinuities has been expressed by Jance (1966a):

"While factor analysis does not, in itself, delimit groups, it does present data in a far more comprehensible form as a basis for the establishment of such groupings by other means."

As stated earlier, the purpose of this investigation was not to delimit groups but to illustrate the nature of variation within existing groups. The results of the principal component ordinations that follow should be interpreted in the light of these qualifications.

### 3.5 Results of the numerical analyses

### 3.5.1 Preamble

The results of the first analysis suggested reasons for conducting several slightly different analyses (see page 61). Therefore, the results of each analysis will be presented in the sequence in which they were performed and the purpose of each will be explained before the results are considered. For each analysis, tables are given which present the most significant correlation coefficients, the eigenvalues for all of the components extracted, and the factor loadings for those characters contributing the most variation to each component extracted. Characters and samples will be referred to at times by the code numbers listed in tables 3.2 and 3.4.

### 3.5.2 Analysis 1

A matrix of correlation coefficients was calculated as the first stage in the analysis (this was performed as part of the computer programme). The matrix was computed

from 41 characters in each of the 211 samples in the data matrix.

### Correlation coefficients

Where <u>n</u> = the number of samples, the significance of a correlation coefficient can be determined by comparison with tabulated critical values, entered at <u>n-2</u> degrees of freedom (Snedecor and Cochran, 1968 section 7.6 and table A.11). In this matrix, with 211 samples, the critical values were 0.14 at the 5% level of probability and 0.18 at the 1% level. A comparison of the observed values with critical values revealed correlations between many characters. The 41 by 41 correlation matrix is not reproduced in full, but those character pairs with correlation coefficients of absolute value greater than 0.5 are listed in table 3.5.

An initial examination of the correlation matrix may reveal weaknesses in some of the characters chosen for inclusion. Consideration must be given to the degree of correlation between characters and also the nature of those characters. In particular, attention should be given to characters, such as ratios, that are derived from other characters that are also included.

TABLE 3.5

CORRELATION COEFFICIENTS WITH ABSOLUTE VALUES GREATER THAN 0.59 IN THE CORRELATION MATRIX DESCRIBING RELATIONSHIPS BETWEEN 41 CHARACTERS IN TERMS OF 211 SAMPLES.

Characters (described by code)		
35 & 37	Length of third lateral and length of fifth lateral	0.90
25 & 27	Length of bract and ratio utricle length:bract length	-0.88
33 & 35	Length of first lateral and length of third lateral	0.88
36 & 38	Width of third lateral <u>and</u> width of fifth lateral	0.87
33 & 37	Length of first lateral <u>and</u> length of fifth lateral	0.82
34 & 36	Width of first lateral and width of third lateral	0.82
34 & 38	Width of first lateral <u>and</u> width of fifth lateral	0.81
8 & 10	Seed length and greatest width of seed	0.80
21 & 24	Variance of character 9 and variance of character 12	0.80
29 & 35	Terminal spike length and length of third lateral	0.79
2 & 11	Size of utricle cap and ratio utricle cup:cap	<b>-</b> 0.78
1 & 11	Size of utricle cup <u>and</u> ratio utricle cup:cap	0.77
39 & 40	Relative length of first lateral and relative length of fifth lateral	-0.77
4 & 16	Number of tepals <u>and</u> variance of the number of tepals	-0.76

Characters (described by code)	Description of Characters (for full description see table 3.4 p.	Correlation <b>35</b> )Coefficient
8 & 12	Seed length and ratio seed width: seed length	-0.75
29 & 33	Terminal spike length and length of first lateral	0.74
1 & 25	Size of utricle cup and bract length	0.72
5 & 28	Tepals with excurrent midribs and ratio longest tepal:bract length	0.72
31 & 36	Terminal spike width at 1 cm and width of third lateral	0.68
2 & 28	Size of utricle cap and ratio longest tepal:bract length	0.67
31 & 38	Terminal spike width at 1 cm and width of fifth lateral	0.67
27 & 28	Ratio utricle length:bract length and ratio longest tepal:bract length	0.65
5 & 11	Tepals with excurrent midrib and ratio utricle cup:cap	-0.64
4 & 5	Number of tepals <u>and</u> tepals with excurrent midrib	0.63
2 & 5	Utricle cap size <u>and</u> tepals with excurrent midrib	0.62
11 & 28	Ratio utricle cup:cap and ratio longest tepal:bract	0.62
<b>31</b> & 34	Terminal spike width at 1 cm and width of first lateral	0.62
25 & 36	Bract length <u>and</u> width of third lateral	0.60

Some of the highest correlations observed in this analysis were between the lengths of the first, third and fifth lateral branches of the terminal inflorescence.

This situation is not unexpected and doubtless reflects the degree to which these characters are under common genetic and environmental control. These characters were correlated at a slightly lower level with the length of the terminal spike (i.e. that part of the terminal inflorescence above the first lateral) and thus all of them reflect changes in the size of the inflorescence. The characters that described the width of the lateral branches were not correlated with the lengths but were correlated among themselves, again suggesting a common control. It did not seem reasonable to include two sets of three characters that were apparently measures of just two basic functions. Thus in further analyses some of these characters were omitted.

The high correlation between the length and greatest width of the seed indicated that the ratio of these two characters varied very little. The negative correlation between this ratio and the length of the seed indicated that the small amount of variation that existed in this ratio resulted from changes in the length of the seed that were not correlated with changes in the greatest width. In these circumstances this ratio supplied little further information concerning the variation in the matrix and at the same time duplicated the information provided by the characters seed length and seed width. In subsequent analyses the ratio of seed width to seed length was omitted.

A similar situation occurred with the characters

describing the size of the utricle "cup", size of the utricle "cap" and the ratio of the two. The ratio appeared merely to duplicate information describing changes in the sizes of "cup" and "cap". This ratio was also excluded from the characters used in further analyses.

The negative correlation between the mean number of tepals (per flower) and the variance (per plant) of the number of tepals illustrated a different situation. The variance was low when the tepal number was at its highest and with lower tepal numbers the variance was higher. This suggests that a low mean tepal number represents a loss of tepals from a basic 5-merous condition. For the purposes of this analysis, the correlation between these two characters meant that they were supplying the same information concerning the state of the tepals. Consequently one of the characters There were no strong was omitted from subsequent analyses. reasons for choosing between the characters, and arbitrarily it was decided that the variance character would be excluded.

The negative correlation between the length of the bract and the ratio of utricle length to bract length indicated that most of the variation in the latter character could be explained in terms of variation in the bract length. Both of these characters were retained since they had been used by several authors as diagnostic characters between these species (Table 3.3). Furthermore, length of the utricle was not included as a separate character. Despite the high

correlation between the characters, it will be seen in analyses to be described that their contributions to the principal component axes were often quite different in magnitude. In analysis 2, the two characters contributed large factor loadings to different axes.

Where the two characters contributed large amounts to the same component, this might have had the effect of increasing the apparent variation in the ordinated samples. Thus any discontinuities that were observed would be accentuated by the exclusion of one of the characters.

The high correlation between the length of the utricle cup and the length of bract appeared to reflect a trend in general size. Both of these characters were correlated at lower levels with other characters that reflected size; for example tepal length, seed length, length of the terminal spike, mid-length width of the terminal spike, and the widths of the first, third and fifth laterals. The negative correlation between the <u>relative</u> lengths of the first and fifth laterals was a demonstration that apical dominance declined with distance from the apex.

# Factor analysis

In the light of the observed correlations that revealed undesirable redundancy of information, the analysis was terminated and a new correlation matrix was computed and analysed. This is described as Analysis 2.

# 3.5.3 Analysis 2

A new correlation matrix was prepared excluding the

following characters: the ratio utricle cup/utricle cap (character 11); the ratio seed width/seed length (character 12); the variance of the number of tepals (character 16); the variance of the utricle cup/cap ratio (character 23); the variance of the seed width/length ratio (character 24); the lengths of the first and fifth laterals (characters 33 and 37 respectively); and the widths of the first and fifth laterals (characters 34 and 38 respectively).

# Correlation coefficients

The values of the coefficients between characters in the new matrix were identical to those in the initial matrix for those characters included. The dimensions of the new matrix were 32 by 32.

# Factor analysis

An R-type analysis was performed upon the new correlation matrix. Components were extracted until the first with an eigenvalue less than unity was encountered. The analysis was terminated at this point.

# <u>Eigenvalues</u>

Eigen values for the first eight component axes, and the proportion of variance accounted for by each component are presented in table 3.6. Over 45% of the variance in the data was accounted for by the first three components extracted, while the first eight components together accounted for almost 70%.

TABLE 3.6

# EIGENVALUES FOR THE FIRST EIGHT COMPONENT AXES EXTRACTED IN ANALYSIS 2, AND THE PERCENTAGE OF THE TOTAL VARIANCE THAT EACH DESCRIBES

Component: 1 2 3 4 5 6 7 8

Eigenvalue: 5.91 4.79 4.00 1.92 1.48 1.29 1.14 1.10

Percentage of the total variance: 18.9 15.3 12.8 6.14 4.73 4.11 3.62 3.52

### Factor Matrix

Table 3.7 presents a list of those characters with the greatest factor loadings on each of the eight components extracted.

The characters with large factor loadings on the first component axis were mostly characters describing differences in size among the plants sampled. These characters were identified in the correlation matrix by their high coefficients. An exception was character 26 which described the variance of the length of the bract. Character 41, the number of lateral branches in 10 cm below the terminal spike, can also be considered as part of the trend in size (as observed in the correlation matrix, page 50). This character made a negative contribution to the first component axis, which is appropriate, since a lower density of laterals reflects a greater internode length.

TABLE 3.7

FACTOR LOADINGS FOR THOSE CHARACTERS THAT CONTRIBUTE THE MOST VARIATION TO EACH OF THE COMPONENTS EXTRACTED.

Component	Character contributing to the component	Factor loading
1	1 - Size of the utricle cup	0.77
	25 - Length of the bract	0.74
	8 - Seed length	0.68
	29 - Length of terminal spike	0.67
: <u>;</u>	36 - Width of third lateral	0.65
	41 - Number of laterals in 10 cm	-0.60
2	6 - Proportion of tepals with retuse apices	0.60
	27 - Ratio utricle length:bract length	-0.57
	3 - Number of style branches	0.55
	10 - Greatest width of seed	-0.54
	2 - Size of utricle cap	-0.53
	28 - Ratio longest tepal:bract length	-0.51
	30 - Proportion of the length of terminal spike with visible stem	-0.51
	31 - Width of terminal spike, 1 cm from apex	0.51
3	7 - Length of longest tepal	0.82
	4 - Number of tepals	0.71
	5 - Proportion of tepals with excurrent midribs	0.71
	28 - Ratio longest tepal:bract length	0.64
	2 - Size of utricle cap	0.61
	6 - Proportion of tepals with retuse apices	0.61

Component	Character contributing to the component	Factor loading
4	39 - Relative length of first lateral	-0.52
	40 - Relative length of fifth lateral	0.50
5	39 - Relative length of first lateral	0.63
	40 - Relative length of fifth lateral	-0.60
6	3 - Number of style branches	-0.56
7	21 - Variance of the relative distance from the micropyle to point of greatest seed width	-0.49
8	22 - Variance of the greatest width of seed	0.69
	14 - Variance of the size of the utricle cap	-0.51

Most of the large factor loadings on the second component came from characters that are unrelated. Another feature of this component was that no single character or group of characters contributed exceptionally large amounts of variation.

Much of the variance contributed to the third component axis was received from characters describing the state of the female perianth (characters 4,5,6 and 28). Several of the characters that contributed to this axis had already made contributions to a previous axis. This indicates that the variation they described could be partitioned in different directions that were correlated with the direction of variation in different sets of other characters.

Component four described just over 6% of the total variation and subsequent components each described less than 5%. The characters that contributed the most variation to each of these components are listed in table 3.7.

#### Factor scores

In figure 3.2, each of the samples has been ordinated on the first two component axes, using the factor scores as coordinates. In figure 3.3, the samples have been ordinated on the first and third component axes.

In the ordination on axes one and two, samples appear to be concentrated in three zones. By far the largest number of samples form an elongated ellipse with its centre close to the mid-point on axis one and to the right of the mid-point on axis two. This group of samples includes most of those assigned to Amaranthus powellii 'type A' and all of those assigned to A. retroflexus. A second zone occupies an extreme negative position on axis one and a mid-position on axis two. This group consists exclusively of those samples assigned to A. hybridus. The third zone is centered below the mid-point on axis one and at a mid-negative position on axis two. This group consists of samples assigned to A. powellii, with most of them belonging to 'type B'.

Before examining the ordination on axes one and three, it is important to point out that most of the variation described by this ordination has already been described by

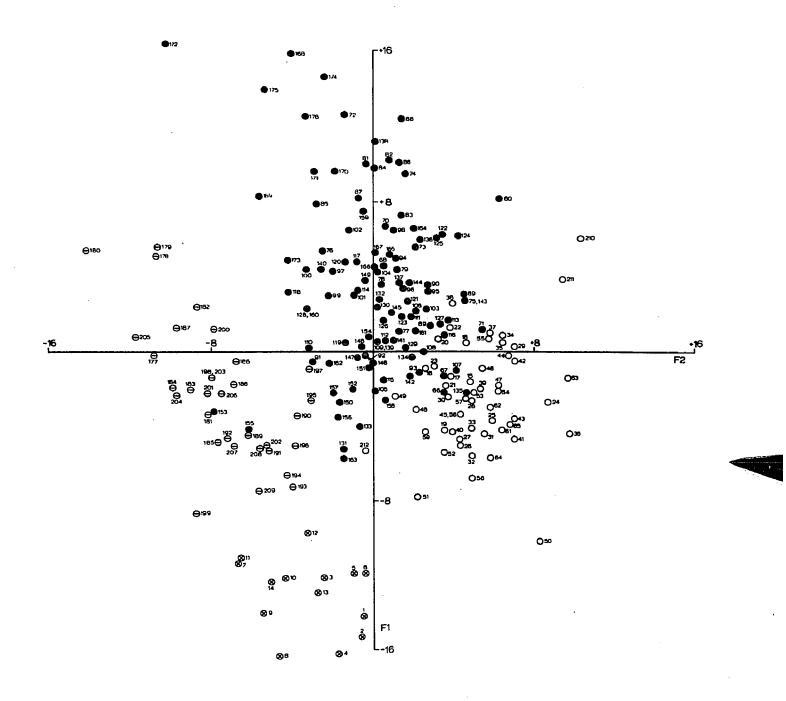


Fig. 3.2 An ordination of each sample on the first two component axes extracted in analysis 2. (a key to the symbols is given in Fig. 3.3)

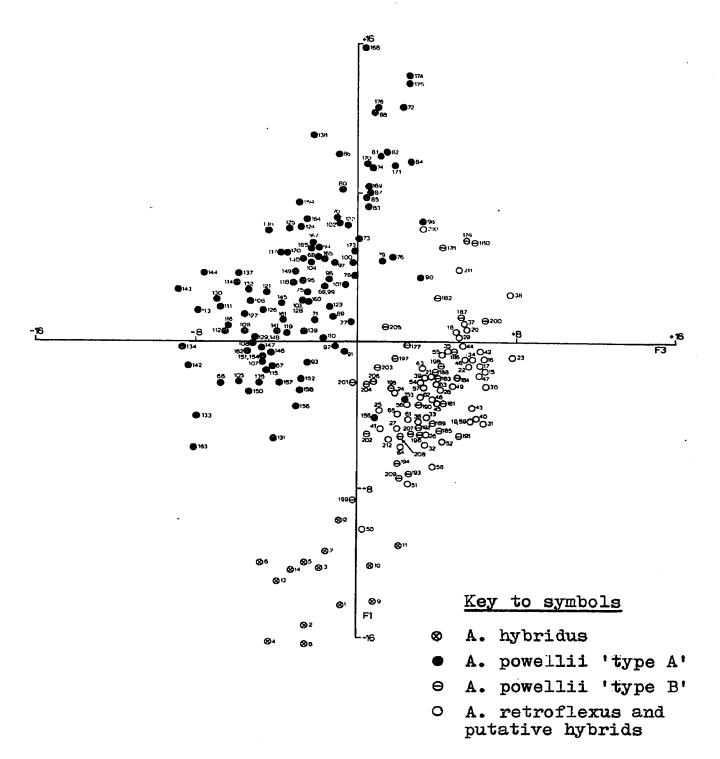


Fig. 3.3 An ordination of each sample on the first and third axes extracted in analysis 2.

the ordination on axes one and two. This is because the variation on axis one is included in both ordinations. Thus any discontinuities in the variation between the samples on axis one will be evident in both ordinations. The value in considering both of these subsequent axes with axis one can be seen in this example.

In the ordination on axes one and three there are again three zones in which the samples are concentrated. The first of these, containing the fewest samples, occupies the extreme negative position on axis one. This group is only distinct from other samples on axis one and it is the same group that can be seen in a similar position in the ordination on axes one and two. The samples included in this group were all assigned to A. hybridus. The two remaining zones are separated partly on axis one and partly The zone that lies predominantly in the on axis three. upper left quadrant of the ordination consists exclusively of samples assigned to A. powellii 'type A'. The other zone, which is predominantly in the lower right quadrant, consists mainly of samples assigned to A. retroflexus and A. powellii 'type B'.

When the position of the samples on each of the three axes are considered together, each of the four presupposed taxa can be distinguished. On axes one and two.

A. retroflexus is indistinguishable from A. powellii 'type A', but is clearly distinguished from 'type B'. On axes one and

three the opposite situation holds; <u>A. retroflexus</u> is distinguished from 'type A' of <u>A. powellii</u> but not from 'type B'.

The distinction between the two 'types' of A. powellii is evident in the ordination on axes one and two, with the exception of two samples of 'type A' which occupy a position removed from other samples of 'type A' and within the group of samples of 'type B'. When this anomaly was noticed, reference was made to the original data scores for the two samples in question (numbers 153 and 155). It was found that at the time the measurements had been made, difficulty had been encountered in assigning these samples to an appropriate 'type'. In most of the characters that were measured these samples had values that resembled 'type B', but in two particular characters on the basis of which other samples had been allocated, they resembled 'type A'. The two characters in question were the number of tepals, which in most flowers was four, and the ease with which the utricle dehisced (a subjective value, not included as a character in the analysis), in this case with some As a result these two samples, together with resistance. another from the same source (number 154), were tentatively assigned to 'type A'. In the light of this analysis, this allocation was erroneous for samples 153 and 155. 154 differed morphologically from the other two samples and was correctly assigned to 'type A'.

Table 3.8 indicates which of the component axes must be considered in order to differentiate between any pair of taxa.

#### TABLE 3.8

A PAIRED COMPARISON OF TAXA IN TERMS OF THE COMPONENT AXES THAT MUST BE CONSIDERED IN ORDER TO DISTINGUISH BETWEEN THE TAXA.

	A. hybridus	A. powellii 'type A'	A. powellii 'type B'
A. powellii 'type A'	1		
A. powellii 'type B'	1	1 and 2	
A. retroflexus	1 or 2	1 and 3	2

A further point of interest that is revealed by the two ordinations is the difference in the degree of variation within each taxon. The samples of A. hybridus and A. retroflexus present tighter clusters and are less variable than either 'type' of A. powellii. This may be explained in part by the fact that more samples were included of A. powellii 'type A' than of any other taxa. However there are fewer samples that represent A. powellii 'type B' than samples that represent A. retroflexus yet variation in the former exceeds that in the latter.

Much of the extreme variation in both 'types' of

A. powellii is contributed by those samples that represent
plants grown at the experimental farm, an environment that



allowed optimal development. With some of these samples a comparison can be made with samples that represent field collections from the original locality at which seeds of the experimental plants were collected (see table 3.2). Comparable samples are distant from each other in the ordination and this suggests that much of the variation observed has resulted in response to environmental differences. If this is so, it is interesting that environmental influences are pronounced in only one of the three species.

Finally, the position of the three samples described as putative hybrids should receive comment. In the first ordination (axes 1 and 2), samples 210 and 211 occupy distinct positions while sample 212 occupies a position peripheral to <u>A. powellii</u> 'type A' but it is also about mid-way between the clusters of the other three taxa. In the ordination on axes one and three, these samples come to lie closer to, but hardly within, clusters of the recognised taxa.

# Implications for further analysis

Although this analysis clearly demonstrated the distinctness of each of the three species and the discontinuous variation within A. powellii, the suggestion also was made that much of the variation arose from environmental influences. Two modifications of the analysis were envisaged as means of isolating the variation suspected to be of environmental origin. The first of these

was to remove those characters that were suspected of exhibiting the greatest environmental influence, and the second modification was to partition the samples into groups representative of the three major environments included; the experimental farm, the greenhouse, and natural sites. These modifications were made in the analyses described in the following pages.

#### 3.5.4 Analysis 3

As can be seen in table 3.25, those characters that described the morphology of the inflorescence (characters 29 to 41) were more variable than the other characters. This observation confirmed the impression gained while handling the material that these characters were the least reliable for use in assigning samples to taxa. Consequently, the inflorescence characters were omitted and a new analysis was performed. In addition those characters excluded from analysis 2 were excluded from this analysis and all further analyses.

# Correlation coefficients

The number of samples included in this analysis was the same as in analyses 1 and 2. Therefore the correlation coefficients between those characters included were of the same value as in analysis 1. Characters with large absolute correlation coefficients are listed in table 3.5, p. 46. It should be remembered that some of the characters listed in table 3.5 were not included in this analysis.

### Factor analysis

An R-type analysis was performed upon the new correlation matrix. Components were extracted until the first with an eigenvalue less than unity was encountered.

#### Eigenvalues

Eigenvalues for the first seven components are listed in table 3.9. In total, these accounted for 71.5% of the variation in the data set, while the first three axes accounted for 49% of the variation. The first three axes in this analysis accounted for about the same amount of variation as the first three axes in analysis 2.

TABLE 3.9

# EIGENVALUES FOR THE FIRST SEVEN COMPONENT AXES EXTRACTED IN ANALYSIS 3, AND THE PERCENTAGE OF THE TOTAL VARIANCE THAT EACH DESCRIBES

Component:	1	2	3	4	5	6	7
Eigenvalue:	4.79	3.20	3.06	1.57	1.26	1.10	1.03
Percentage of the total variance:	21.4	14.3	13.7	7.02	5.61	4.88	4.60

# Factor Matrix

Factor loadings for each character on each of the first seven components are presented in table 3.10.

The characters that contributed large amounts of variation to component axis one included some that had contributed to axis one in the previous analysis and others that had contributed to axis two. This indicated that with the

inflorescence characters removed, there had been a change in the direction of major variation with respect to the characters remaining. In particular, the tendency of component one to describe differences in size was absent in this analysis.

TABLE 3.10

FACTOR LOADINGS FOR THOSE CHARACTERS THAT CONTRIBUTE THE MOST VARIATION TO EACH OF THE COMPONENTS EXTRACTED IN ANALYSIS 3.

Component	Character contibuting to the component	Factor loading
1	28 - Ratio longest tepal:bract length	-0.82
	5 - Proportion of tepals with excurrent midrib	-0.76
	27 - Ratio utricle length:bract length	-0.76
	25 - Length of bract	0.74
	1 - Size of utricle cup	0.71
	2 - Size of utricle cap	-0.65
	4 - Number of tepals	-0.61
2	8 - Length of seed	0.82
	10 - Greatest width of seed	0.78
	6 - Proportion of tepals with retuse apices	-0.72
	18 - Variance of the proportion of tepals with retuse apices	-0.63
3	7 - Length of the longest tepal	0.95
4	20 - Variance of the seed length	0.63
5	3 - Number of style branches	-0.48
	15 - Variance of the number of style branches	0.48
6	22 - Variance of the greatest width of the seed	0.82
7	21 - Variance of the relative distance between the micropyle and point of greatest seed width	0.53

Component two also reflected the change in the major direction of variation. Characters that contributed to this component included some that had contributed to component one and others that had contributed to component two in the previous analysis.

Component three received a large contribution from one character, the length of the longest tepal, and only small amounts of variance from other characters. Contributions to the remaining components extracted are presented in table 3.10.

#### Factor scores

In figure 3.4, each of the samples has been ordinated on the first two component axes, using factor scores as coordinates. In figure 3.5, histograms represent the frequency distribution of each of the predetermined taxa on component axis three. In order to obtain the frequency distribution, variation on the third axis was described within classes of 0.99 units interval. Interpretation of the distribution of samples on axis three in this analysis did not depend on knowledge of the distribution on axis one. For this reason the frequency distribution on one axis was chosen (as opposed to an ordination on two axes) as the best illustration of the distribution.

Three main groups can be detected in the ordination of samples on the first two component axes. The clustering within these groups appears tighter than in the ordination in analysis 2. Samples representing A. hybridus do not

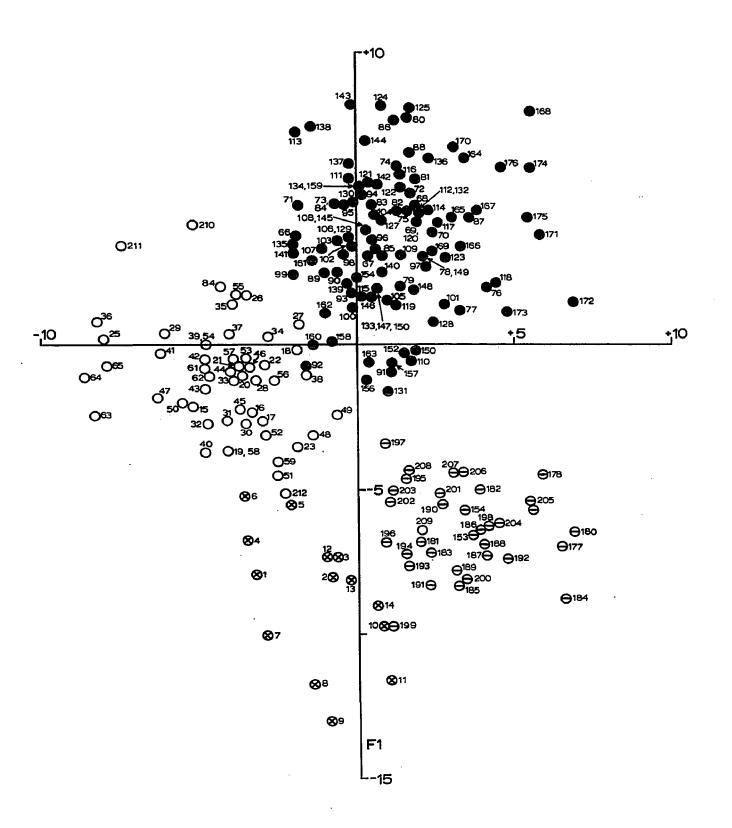
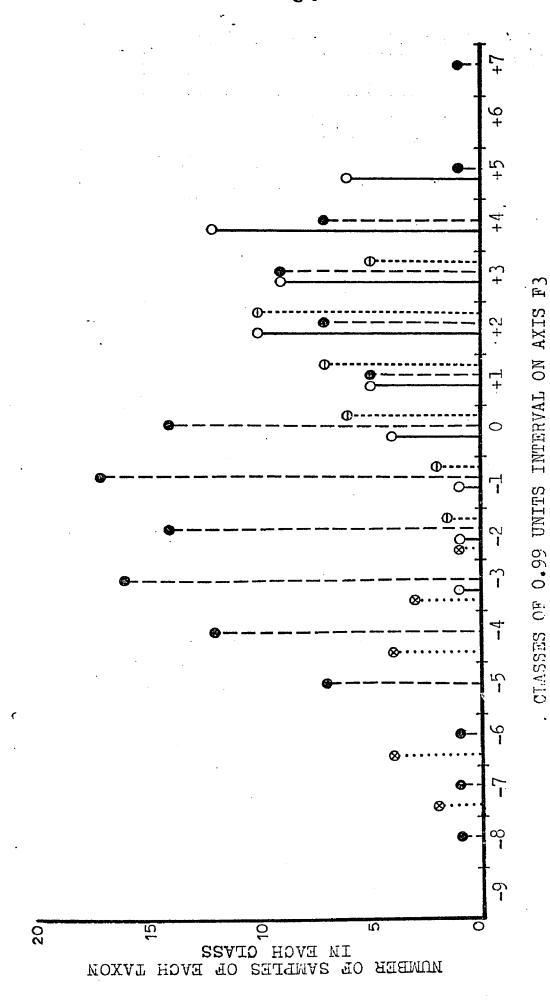


Fig. 3.4 An ordination of each sample on the first two axes extracted in analysis 3.

(a key to symbols is given in Fig. 3.3)



The frequnecy distribution of samples of each taxon on the third component axis extracted in analysis 3 3.5 Fig.

A. retroflexus 11: type B' Ħ Taxa may be identified as follows: A. hybridus <sup>2</sup> 11 No te

form as distinct a group in this analysis as in analysis 2, suggesting that the inflorescence characters that were excluded may have aided in the segregation of this species. The amount of variation within the cluster of samples of each taxa appears to be proportional to the number of samples.

Table 3.11 indicates which of the component axes must be considered in order to differentiate between taxa.

#### TABLE 3.11

A PAIRED COMPARISON OF TAXA IN TERMS OF THE COMPONENT AXES THAT MUST BE CONSIDERED IN ORDER TO DISTINGUISH BETWEEN THE TAXA.

	A. hybridus	A. powellii 'type A'	A. powellii 'type B'
A. powellii 'type A'	1		
A. powellii 'type B'	3 or 1 & 2	1	
A. retroflexus	1 or 3	1 & 2	2

In signifying the presupposed identity of the samples in this ordination, samples 153 and 155 have been described as A. powellii 'type B' for the reasons given in analysis 2, page 59. The position of these samples in this ordination strengthens the view that they were originally determined incorrectly.

# 3.5.5 Analysis 4

To complement analysis 3, a correlation matrix was

prepared including only those characters that were included in analysis 2 and excluded from analysis 3. This then was a matrix of inflorescence characters. A factor analysis was performed upon this matrix in order to investigate the hypothesis that variation in inflorescence characters served to obscure taxonomic distinctions.

### Correlation coefficients

The values in the reduced correlation matrix were the same, for those characters included, as the values in the matrix described in analysis 1.

### Factor analysis and eigenvalues

An R-type factor analysis was performed upon the 9 by 9 correlation matrix, and three components were extracted with eigenvalues greater than unity. The eigenvalues for these components were, in the order that they were extracted: 3.35, 2.18 and 1.37. Together these three components accounted for 78% of the total variation.

# Factor loadings

Characters that contributed large amounts of variation to the three component axes are listed in table 3.12.

The first component accounted for 38% of the total variation and consisted of characters reflecting the overall size of the plant they represented. Two of these characters had contributed to the first axis in analysis 2.

When factor loadings on the first, second and third axes are considered it can be seen that all of the original characters have contributed large amounts of variation to one or other of these axes.

TABLE 3.12

FACTOR LOADINGS FOR THOSE CHARACTERS THAT CONTRIBUTE THE MOST VARIATION TO EACH OF THE COMPONENTS EXTRACTED.

Component	Character contributing to the component	Factor loading
1	29 - Length of the terminal spike	0.86
	35 - Length of the third lateral	0.84
	41 - Number of laterals in 10 cm.	-0.78
	30 - Proportion of terminal spike in which stem is visible	0.61
2	31 - Width of terminal spike at 1 cm.	0.83
	36 - Width of third lateral	0.72
	32 - Width of the terminal spike at mid-length	0.69
3	40 - Relative length of the fifth lateral	-0.71
	39 - Relative length of the first lateral	0.66

## Factor scores

In figure 3.6, the samples are ordinated according to their factor scores on the first two axes.

No separate groups can be distinguished in this ordination. Although the different taxa show some segregation within the one group of samples, there is much overlap in the distribution of each. The factor scores on the third component have been inspected and reveal no meaningful discontinuities. Thus a graphical representation of distribution on the third axis has not been included.

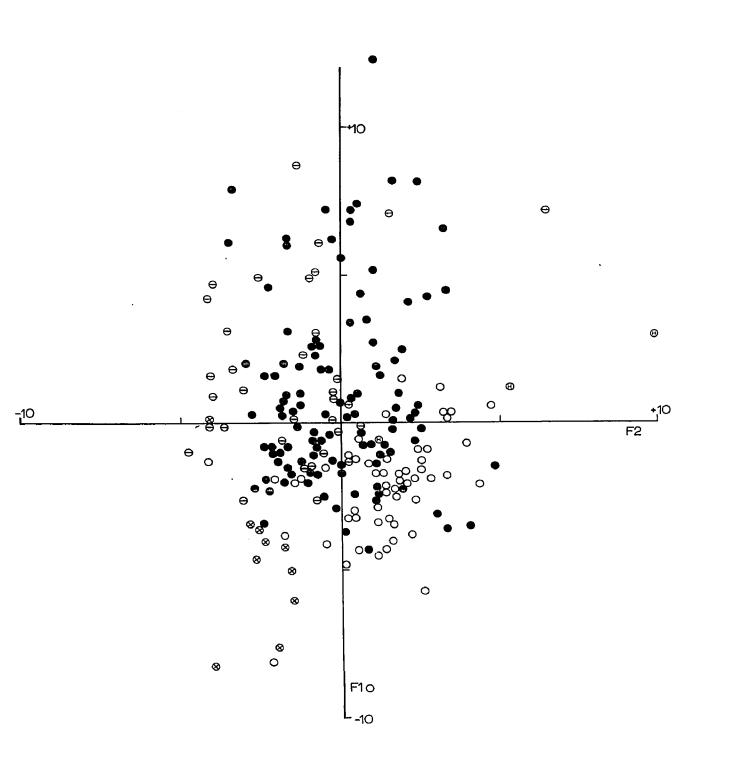


Fig. 3.6 An ordination of each sample on the first two axes extracted in analysis 4.

(a key to the symbols is given in Fig. 3.3)

#### 3.5.6 Analysis 5

The second modification suggested from the results of analysis 2, page 61 was a partitioning of samples from different types of environments. This was achieved by analysing separately samples representing plants from original field sites, from the experimental farm and from the greenhouse. In this analysis, 95 samples were included which represented plants collected in the various field sites. All of the characters included in analysis 2 were included in this analysis.

#### Correlation coefficients

Correlation coefficients describe character relationships as they are expressed in the samples. Since the number of samples included in this analysis differed from the number in analyses 1 and 2, it was necessary to calculate new coefficients. Critical values for the significance of coefficients in this analysis were 0.20 and 0.26 at the 5% and 1% levels of probability respectively. Table 3.13, lists those characters with correlation coefficients greater than 0.59 (absolute value).

# Factor analysis and eigenvalues

An R-type analysis was performed upon the new correlation matrix and eight components were extracted with eigenvalues greater than unity. The eigenvalues for each component extracted are listed in table 3.14.

TABLE 3.13

# CORRELATION COEFFICIENTS WITH ABSOLUTE VALUES GREATER THAN 0.59 IN THE CORRELATION MATRIX OF ANALYSIS 5.

LIMI O.	)) IN IIII COMMINATION MAININ OF ANADIDID )	•
Coded characters	Description of Characters (for full description see table 3.4 p. )	Correlation Coefficient
25 & 27	Length of bract and ratio utricle length: bract length	-0.89
8 & 10	Seed length and greatest width of seed	0.86
29 & 35	Length of the terminal spike and length of third lateral	0.85
39 & 40	Relative length of first lateral and relative length of fifth lateral	-0.77
32 & 36	Width of terminal spike at mid-length and width of third lateral	0.76
29 & 30	Length of terminal spike <u>and</u> proportion of terminal spike with visible stem	0.74
31 & 32	Width of terminal spike at 1 cm and width of terminal spike at mid-length	0.74
25 & 32	Length of bract <u>and</u> width of terminal spike at mid-lengh	0.71
8 & 41	Seed length <u>and</u> number of laterals in 10 cm	-0.69
25 & 28	Length of bract and ratio longest tepal:bract length	-0.69
5 & 28	Tepals with excurrent midribs and ratio longest tepal:bract length	0.69
1 & 25 <sup>5</sup>	Size of utricle cup and length of bract	0.68
6 & 8 .	Tepals with retuse apices and seed length	0.66
25 & 36	Length of bract <u>and</u> width of the third lateral	0.66
29 & 41	Length of terminal spike <u>and</u> number of laterals in 10 cm	-0.65
2 & 28	Size of utricle cap <u>and</u> ratio longesttepal:bract length	0.62
8 & 29	Seed length <u>and</u> length of terminal spike	0.61

#### TABLE 3.14

# EIGENVALUES FOR THE FIRST EIGHT COMPONENT AXES EXTRACTED IN ANALYSIS 5, AND THE PERCENTAGE OF THE TOTAL VARIANCE THAT EACH DESCRIBES

Component:	1	2	3	4	5	6	7	8
Eigenvalue:	6.95	5.04	3.36	2.18	1.56	1.37	1.21	1.12
Percentage of total variance:	22.2	16.1	10.8	6.96	4.98	4.39	<b>3.</b> 86	3 <b>•</b> 59

#### Factor loadings

The characters listed in table 3.15 made the greatest contributions to each of the component axes extracted.

Component one received large contributions from several characters, the majority of which could be considered to reflect size differences. Component two included large contributions from the two major seed characters, while component three received a large contribution from only one character, the length of the longest tepal.

# Factor scores

In figure 3.7 each sample has been ordinated on axes one and two using factor scores as coordinates. Figure 3.8 presents a frequency distribution of samples within classes on axis three.

Only two samples representing A. hybridus were available for inclusion in this analysis, (several other specimens collected in the field were too immature to provide all of the necessary measurements) and therefore it is hardly appropriate to discuss the position of this species in the

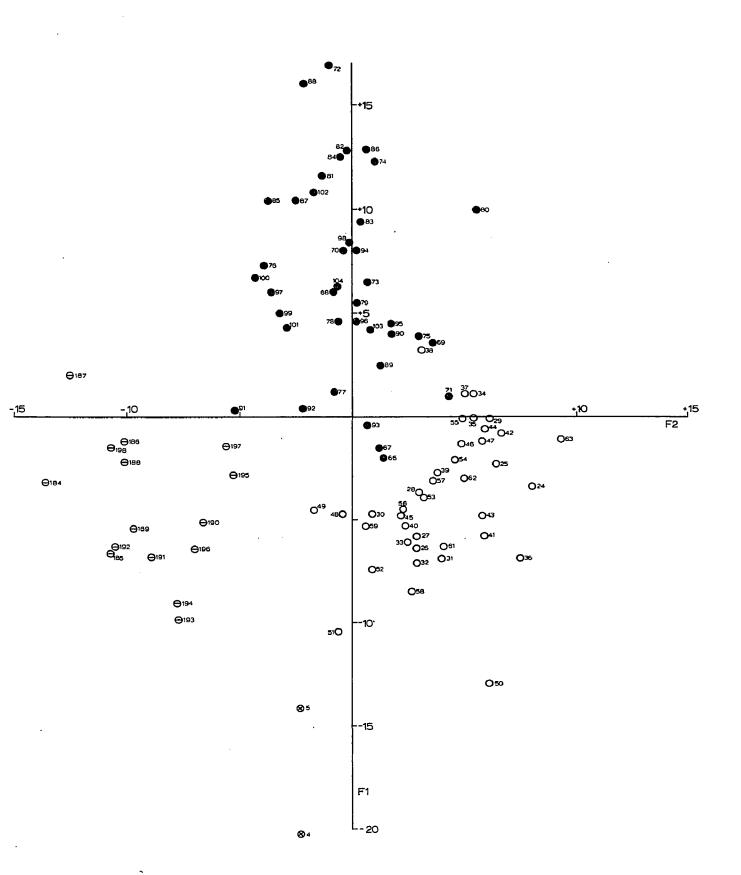
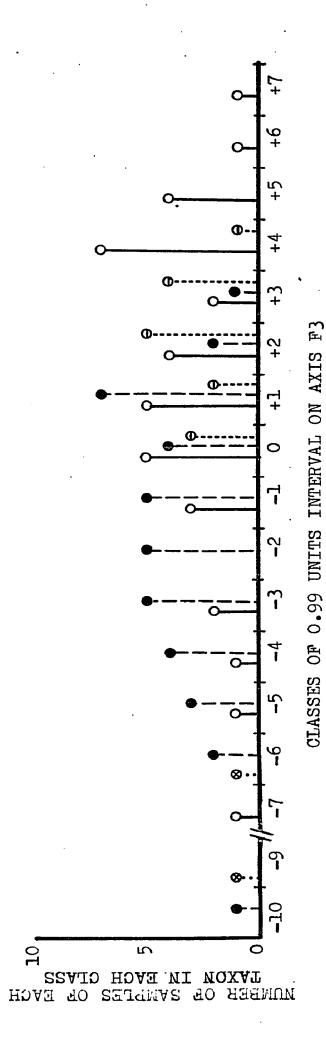


Fig. 3.7 An ordination on the first two axes of analysis 5 of those samples representing plants collected in the field.

(a key to the symbols is given in Fig. 3.3)



The frequency distribution of samples of each taxon on the third component axis extracted in analysis 5. Fig. 3.8

The symbols used to identify taxa are described in Fig. 3.5, page  $m{67}$ No te:

ordination. Samples of the three other taxa are reasonably distinct although none of the groups has a concentration of samples at its centre.

**TABLE 3.15** 

# FACTOR LOADINGS FOR THOSE CHARACTERS THAT CONTRIBUTE THE MOST VARIATION TO EACH OF THE COMPONENTS EXTRACTED.

Component	Character contributing to the component	Factor loading
1	29 - Terminal spike length	0.78
	35 - Length of the third lateral	0.75
	36 - Width of the third lateral	0.73
	25 - Bract length	0.72
	1 - Size of the utricle cup	0.64
	30 - Proportion of length of terminal spike with stem visible	0.64
	41 - Number of laterals in 10 cm.	-0.64
	9 - Relative distance from micropyle to point of greatest seed width	0.61
	32 - Width of terminal spike at mid-lengt	h 0.61
2	6 - Proportion of tepals with retuse apices	0.63
	8 - Seed length	-0.63
	10 - Greatest width of the seed	-0.63
3	7 - Length of the longest tepal	0.89
4	39 - Relative length of first lateral	-0.67
	40 - Relative length of fifth lateral	0.67
5	3 - Number of style brandes	-0,52
	22 - Variance of the greatest seed width	0.52

Component	Character contributing to the component	Factor loading
6	21 - Variance of the relative distance from the micropyle to the point of greatest seed width	0.65
	15 - Variance of the number of style branches	-0.52
7	14 - Variance of the size of the utricle cap	0.52
	26 - Variance of the bract length	0.48
8	No character contributed over 0.38 to this component.	

Table 3.16 indicates which components must be considered in order to differentiate between taxa. A. hybridus has been excluded from this comparison because of the low representation of the species in this analysis.

# **TABLE 3.16**

A PAIRED COMPARISON OF TAXA IN THE TERMS OF THE COMPONENTS IN ANALYSIS 5 THAT MUST BE CONSIDERED IN ORDER TO DISTINGUISH BETWEEN THE TAXA.

	A. powellii type_A'	A. powellii type B'	
A. powellii 'type B'	1 & 2 or 1 & 3		
A. retroflexus	1 & 2	2	

# 3.5.7 Analysis 6

Samples representing plants grown in the greenhouse were included in this analysis. The characters that were included

were the same as in analysis 2.

### Correlation coefficients

A new correlation matrix was prepared from the 81 samples included in this analysis. Critical values for significant correlation were 0.22 and 0.28 for the 5% and 1% level of probability respectively. Character pairs with correlation coefficients greater than 0.59 (absolute value) are listed in table 3.17.

TABLE 3.17

CORRELATION COEFFICIENTS WITH ABSOLUTE VALUES GREATER THAN 0.59 IN THE CORRELATION MATRIX OF ANALYSIS 6

Coded characters	Description of characters	Correlation Coefficient
6 & 18	Tepals with retuse apices and variance of tepals with retuse apices	0.94
25 & 27	Length of bract <u>and</u> ratio utricle length:bract length	-0.93
1 & 25	Size of utricle cup <u>and</u> length of bract	0.79
27 & 28	Ratio utricle length:bract length and ratio longest tepal:bract length	-0.75
39 & 40	Relative length of first lateral and relative length of fifth lateral	-0.75
2 & 5	Size of utricle cap <u>and</u> tepals with excurrent midribs	0.73
5 & 28	Tepals with excurrent midrib and ratio longest tepal:bract length	0.70
1 & 27	Size of utricle cup and ratio utricle length:bract length	-0.69
2 & 28	Size of utricle cap and ratio longest tepal:bract length	0.69

Coded characters	Description of characters	Correlation coefficient
1 & 5	Size of utricle cup <u>and</u> tepals with excurrent midrib	<b>-</b> 0.66
8 & 10	Seed length and greatest width of seed	0.66
8 & 9	Seed length <u>and</u> relative distance from micropyle to point of greatest width	0.65
4 & 5	Number of tepals <u>and</u> tepals with excurrent midribs	0.64
1 & 4	Size of utricle cup <u>and</u> number of tepals	0.60
29 & 35	Length of terminal spike and length of third lateral	0.60

### Eigenvalues

Nine component axes with eigenvalues greater than unity were extracted by an R-type factor analysis. These eigenvalues are presented in table 3.18.

#### TABLE 3.18

EIGENVALUES FOR THE FIRST NINE COMPONENTS EXTRACTED IN ANALYSIS 6, AND THE PERCENTAGE OF THE TOTAL VARIANCE DESCRIBED BY EACH

7 8 5 6 3 2 Component: 1

Eigenvalue: 7.46 4.35 2.65 2.43 1.90 1.68 1.45 1.26 1.04

Percentage

of total

23.8 13.8 8.45 7.76 6.06 5.36 4.62 4.02 3.31 variance:

# Factor loadings

The characters that contributed the greatest variation to the first nine components are listed in table 3.19.

TABLE 3.19

FACTOR LOADINGS FOR THOSE CHARACTERS THAT CONTRIBUTE THE MOST VARIATION TO EACH OF THE COMPONENTS EXTRACTED.

Component	Character contributing to the component	Factor loading
1	1 - Size of utricle cup	0.91
	25 - Bract length	0.86
	27 - Ratio utricle length:bract length	-0.84
	28 - Ratio longest tepal:bract length	-0.74
	5 - Proportion of tepals with excurrent midribs	-0.71
	9 - Relative distance from micropyle to point of greatest seed width	0.67
	4 - Number of tepals	0.66
	2 - Size of utricle cap	<b>-</b> 0.65
	17 - Variance of character 5 (above)	0.61
	36 - Width of the third lateral	0.60
2	41 - Number of laterals in 10 cm.	-0.77
	7 - Length of the longest tepal	0.76
	35 - Length of the third lateral	0.71
	10 - Greatest width of seed	0.65
3	6 - Proportion of tepals with retuse apices	0.83
	18 - Variance of character 6 (above)	0.83
4	13 - Variance of the size of utricle cup	0.64
	20 - Variance of seed length	0.64
	21 - Variance of character 9 (above)	0.50
5	3 - Number of style branches	0.61
6	39 - Relative length of first lateral	0.73

Component	Character contributing to the component	Factor loading
7	32 - Width of terminal spike at half- length	0.59
8	22 - Variance of the greatest width of seed	0.45
9	22 - Variance of the greatest width of seed	-0.71
		11

The first component received large contributions from ten characters including several of the 'size' characters noted previously and several of the characters describing the female perianth. Characters describing the overall size of a plant also contributed large values to component two. Component three consisted essentially of one major character, the proportion of tepals with retuse apices. The other character contributing to this axis was the variance of the number of tepals with retuse apices and in this analysis these two characters were highly correlated. Essentially they were contributing the same information.

Several characters that had contributed large amounts of variation to the first three axes in earlier analyses made much smaller contributions to first three axes in this experiment. Most of these characters were inflorescence characters and this situation indicates that there was much less variation in the characters among plants grown in the greenhouse than among plants grown outside.

The greenhouse represents a fairly uniform environment and it is tempting to suggest that the additional variation observed in these characters in plants from other environments was entirely environmentally induced. However it should be noted that plants were cultured in the greenhouse in 10 inch flower-pots that probably imposed limits on root growth and prevented the plants from reaching the dimensions attained in the experimental field. Differences in light quality, light intensity and relative humidity between the greenhouse and field environments may have contributed also to restrict the full expression of inflorescence characteristics by plants grown in the greenhouse.

#### Factor scores

In figure 3.9 each of the samples has been ordinated on the first two component axes, using factor scores for coordinates. In figure 3.10 histograms represent the frequency distribution of samples on axis 3, and in figure 3.11 this representation is repeated for axis 4.

The number of plants that could originally be raised in the greenhouse was limited by considerations of time and space. As a result a preference was given to plants of A. powellii and only two plants of A. retroflexus were included. The two samples of A. retroflexus were included in this analysis since they had been included in previous analyses. However, their presence had a large effect on axis three. This axis serves to separate the two samples of A. retroflexus from all of the other samples, but it

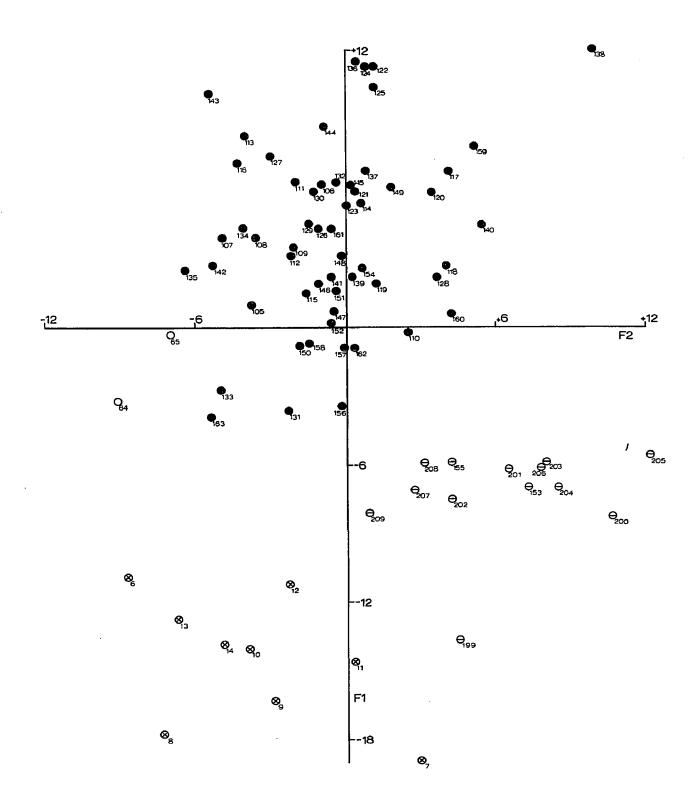
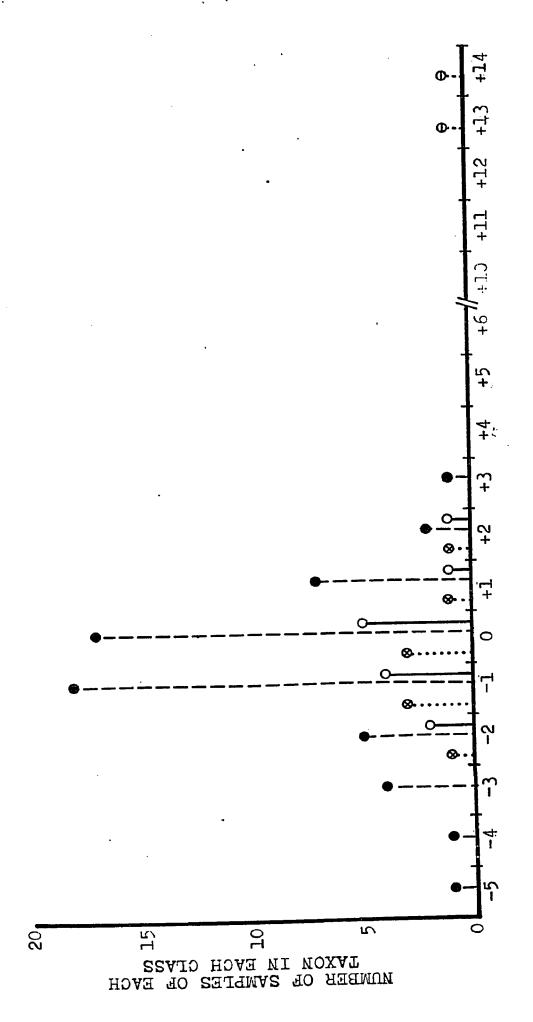


Fig. 3.9 An ordination on the first two axes of analysis 6 of those samples representing plants grown in the greenhouse.

(a key to the symbols is given in Fig. 3.3)



taxon on the The frequency distribution of samples of each third component axis extracted in analysis 6 Fig. 3.10

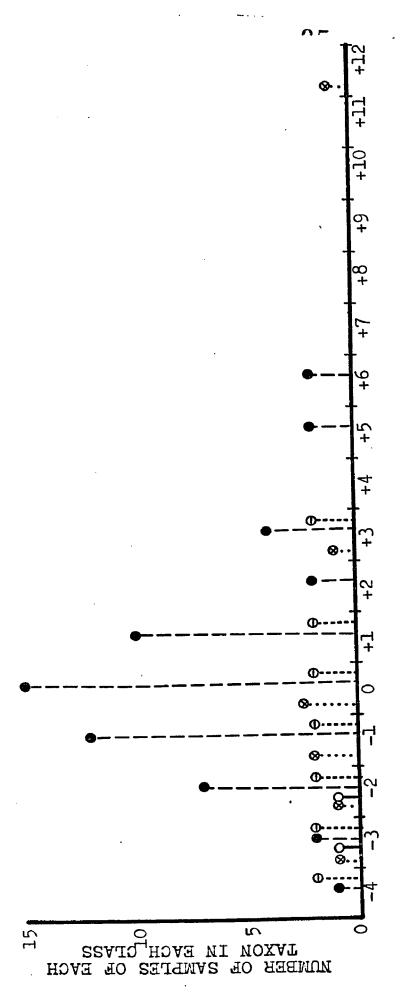
CLASSES OF 0.99 UNITS INTERVAL ON AXIS F3

The symbols used to identify taxa are described in Fig.

Note:

3.5, page 61





The symbols used to identify taxa are described in Fig. 3.5, page 67 The frequency distribution of samples of each taxon on the fourth component axis extracted in analysis 6

Note

CLASSES OF 0.99 UNITS INTERVAL ON AXIS F4

This is not very important, however, because the three taxa are separated clearly on axes one and two. For the sake of completeres, axis four has been illustrated, since the amount of variation described by this axis is almost as much as that described by axis three (Table 3.18).

Table 3.20 indicates which components must be considered in order to differentiate between the taxa.

### TABLE 3.20

A PAIRED COMPARISON OF TAXA IN THE TERMS OF COMPONENTS IN ANALYSIS 6 THAT MUST BE CONSIDERED IN ORDER TO DISTINGUISH BETWEEN THE TAXA.

	A. hybridus	A. powellii 'type A'
A. powellii type A'	1	
A. powellii type B	1 & 2	1 & 2

# 3.5.8 Analysis 7

Data for the seventh analysis consisted of those samples that represented plants grown outside at the experimental farm. These plants are representative of the collections examined in studies of germination (see tables 3.1 and 6.1). One of the characters that had been included in analysis 2 had to be omitted from this analysis, since it occurred with a value of zero in all samples. This character was the variance of the distance between the micropyle to the point of greatest seed width.

# Correlation coefficients

A new correlation matrix was prepared from the 35 samples included in this analysis. Critical values for significant correlation were 0.33 and 0.42 at the 5% and 1% levels of probability respectively.

Character pairs with correlation coefficients greater than 0.59 (absolute value) are listed in table 3.21.

TABLE 3.21

# CORRELATION COEFFICIENTS WITH ABSOLUTE VALUES GREATER THAN 0.59 IN THE CORRELATION MATRIX OF ANALYSIS 7

Coded characters	Description of characters	Correlation coefficient
25 & 27	Length of bract and ratio utricle length:bract length	-0.89
32 & 36	Width of terminal spike at mid-length ar width of third lateral	0.85
29 & 35	Length of terminal spike and length of third lateral	0.84
39 & 40	Relative length of first lateral and relative length of fifth lateral	-0.80
5 & 28	Tepals with excurrent midribs and ratio longest tepal:bract length	0.77
8 & 29	Seed length and length of terminal spil	ke 0.75
9 & 30	Relative distance from micropyle to posof greatest seed width and length of terminal spike with stem vis	
31 & 32	Width of terminal spike at 1 cm and width of terminal spike at mid-length	0.73
8 & 35	Seed length and length of third latera	0.70
5 & 10	Tepals with excurrent midribs and greatest seed width	-0.69

Coded characters		rrelation efficient
30 & 35	Length of terminal spike with stem	
	visible <u>and</u> length of third lateral	0.68
1 & 8	Size of utricle cup and seed length	0.67
2 & 28	Size of utricle cap and ration longest tepal:bract length	0.67
8 & 30	Seed length and length of terminal spike with visible s	tem 0.67
1 & 10	Size of utricle cup <u>and</u> greatest width of seed	0.66
27 & 32	Ratio utricle length:bract length and width of terminal spike at mid-length	-0.65
1 & 25	Size of utricle cup <u>and</u> length of bract	0.64
1 & 30	Size of utricle cup <u>and</u> length of terminal spike with visible s	tem 0.63
1 & 26	Size of utricle cup <u>and</u> variance of bract length	0.62
27 & 31	Ratio utricle length:bract length and width of terminal spike at 1 cm from ap	ex <b>-</b> 0.62
4 & 5	Number of tepals and tepals with excurrent midribs	0.61
31 & 36	Width of terminal spike at 1 cm and width of third lateral.	0.60

# Eigenvalues

The R-type analysis extracted eight components with eigenvalues greater than unity. Together these accounted for 84.2% of the total variation (table 3.22).

### **TABLE 3.22**

EIGENVALUES FOR THE FIRST EIGHT COMPONENT AXES EXTRACTED IN ANALYSIS 7, AND THE PERCENTAGE OF THE TOTAL VARIANCE THAT EACH DESCRIBED

Component:	1	2	3	4	5	6	7	8
Eigenvalue:	7.22	6.58	3.63	2.35	2.08	1.50	1.18	1.02
Percentage of total variance:	23.8	21.7	12.0	7.74	6.86	4.93	3 <b>.</b> 88	3 <b>•</b> 37

### Factor loadings

Factor loadings are listed in table 3.23 for those characters that contributed the most variance to each of the components extracted.

# TABLE 3.23

FACTOR LOADINGS FOR THOSE CHARACTERS THAT CONTRIBUTE THE MOST VARIATION TO EACH OF THE COMPONENTS EXTRACTED.

Component	Character contributing to the component	Factor loading
1	8 - seed length	0.86
	29 - length of the terminal spike	0.86
	30 - proportion of terminal spike with visible stem	0.82
	1 - size of utricle cup	0.81
	10 - greatest width of the seed	0.75
	41 - number of laterals in 10 cm.	-0.74
	35 - length of third lateral	0.73
	26 - variance of the bract length	0.61

Component	Character contributing to the component	Factor loading
2	28 - ratio longest tepal:bract length	0.79
	32 - width of the terminal spike at mid-length	-0.78
	27 - ratio utricle length:bract length	0.76
	2 - size of utricle cap	0.74
	17 - variance of tepals with excurrent midribs	-0.71
	5 - proportion of tepals with excurrent midribs	0.66
	36 - width of third lateral	-0.63
	25 - bract length	-0.62
3	7 - length of the longest tepal	0.64
	4 - number of tepals	0.59
	5 - proportion of tepals with excurrent midribs	0.53
	18 - variance of character 6 (see below)	0.52
	6 - proportion of tepals with retuse apices	0.51
	36 - width of the third lateral	0.50
4	20 - variance of the seed length	0.69
	6 - proportion of tepals with retuse apices	-0.56
	13 - variance of the size of the utricle cup	0.52
5	40 - relative length of fifth lateral	0.82
	39 - relative length of first lateral	-0.74
6	19 - variance of the longest tepal length	h 0.64
7	14 - variance of the size of the utricle cap	0.51

Component	Character contributing to the component	Factor loading
8	9 - relative distance from the micropyle to the point of greatest seed width	-0.43
	2 - size of the utricle cap	-0.41
	19 - variance of the length of the longest tepal.	<b>-</b> 0.40

As in many of the previous analyses, component one consisted of a number of characters concerned with overall plant size. The contributions to components two and three came from a variety of characters that are not obviously related. The first three components were similar in an interesting respect; each of them received large factor loadings from a large number of characters. This is a reflection of the fact that the total variance accounted for by the first three axes was 57%, a high figure compared with previous analyses. Thus plants grown under the uniform conditions of the experimental farm showed greater correlation in their variation, presumably since environmentally induced variation had been minimised.

# Factor scores

Figure 3.12 illustrates the ordination of samples on axes one and two and figure 3.13 presents a frequency distribution of characters on axis three.

Each taxon is very clearly distinguished in the ordination on the first two axes. Although A. hybridus is represented by only three samples, these occupy a distinct position and there is little variation between them.

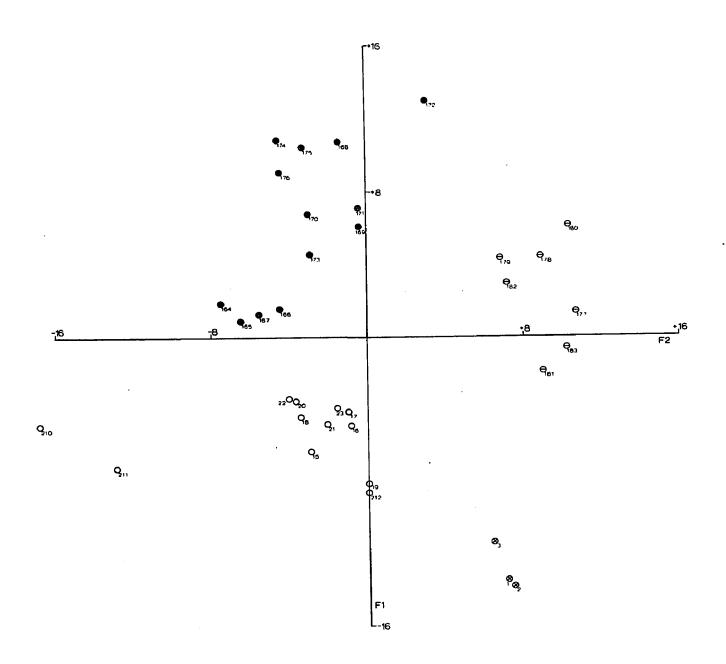
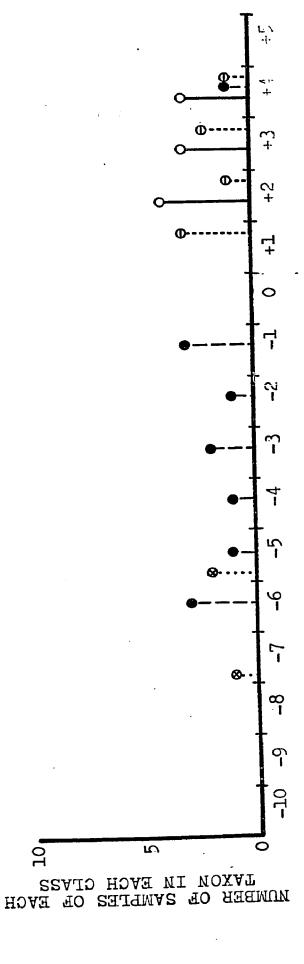


Fig. 3.12 The ordination on the first two axes of analysis 7 of those samples representing plants grown at the experimental farm.

(a key to symbols is given in Fig. 3.3)



The frequency distribution of samples of each taxon on the third component axis extracted in analysis  ${\it 7}$ Fig. 3.13

CLASSES OF 0.99 UNITS INTERVAL ON AXIS F3

are described in Fig. The symbols used to identify taxa Note: The two putative hybrids, samples 210 and 211, occupy a unique position also. The other putative hybrid has a position very close to A. retroflexus.

Within A. powellii 'type A' there is the indication of further differentiation. The samples that represent collection P1 occupy a position that could be considered almost intermediate between the remainder of the group and A. retroflexus. This point is emphasised since the germination response of seeds of collection P1 was found to differ in many ways from the response of seeds of both of the other 'types' of A. powellii (see pages 283 to 285).

Table 3.24 indicates the distinctions between taxa on each of the first three component axes.

TABLE 3.24

A PAIRED COMPARISON OF TAXA IN TERMS OF THE COMPONENTS THAT MUST BE CONSIDERED IN ORDER TO DISTINGUISH BETWEEN THEM.

	A. hybridus	A. powellii 'type A'	A. powellii 'type B'
A. powellii 'type A'	1		
A. powellii 'type B'	1	2 o <b>r</b> 3	
A. retroflexus	1 or 3	1	2

### 3.6 Conclusions

There are several conclusions that can be made in the light of the six component analyses described (analyses 2 to 7).

Both of the techniques that were employed to reduce the influence of environmentally induced variation were successful in improving the separation of taxa in ordinations. When characters were partitioned into those that showed much environmental influence and those that showed very little, two very different ordinations were produced. Ordination of the former characters (analysis 4) revealed no discontinuities in variation and it was not possible to distinguish between taxa. Ordination of the remaining characters (analysis 3) revealed much sharper discontinuities than when both sets of characters were included together.

When the alternative technique was employed and samples were partitioned according to the three major types of environment in which the plants had grown, each analysis resulted in an ordination in which the taxa were clearly distinguished. A comparison of the factor loadings on the major axes in these analyses revealed that inflorescence characters (assumed to be under strong environmental control) were important as discriminants in two of the three environments. There was little variation in inflorescence characteristics in plants grown in the greenhouse. This observation has received comment on page 83.

From these results it is clear that with a careful choice of characters, plants of these taxa taken from diverse environments can be assigned without difficulty to the appropriate taxon.

Several reasons for conducting these analyses were set forth on page 24.

The first reason stated was a need for an efficient description of material used in experimental work. Such a description was provided specifically in analysis 7. At the same time the discontinuities in variation observed in the material included in analysis 7 reflected differences that could be observed in much larger samplings of the species (e.g. analyses 5 and 6).

Within A. powellii discontinuous variation had been suspected and this was clearly demonstrated throughout these analyses. The taxonomic status of the two 'types' of this species is open to speculation. The material studied revealed no individuals that were intermediate between these two 'types' in anything but superficial morphology. This situation occurred despite the fact that of the 13 sites from which 'type B' was collected, 5 also yielded material of 'type A' which was included in this survey. In several of the ordinations it was easier to distinguish between the two 'types' of A. powellii than between either 'type' and A. retroflexus.

The relationship between the germination response of seeds of A. powellii and the morphological identity of the parent plants was mentioned briefly in the results of analysis 7. In studies of germination, collections P3 and P6 have been shown to behave similarly; plants of both these collections fall within 'type B'. Collections P2, P4 and P5 have also been shown to resemble each other in

germination behaviour and plants of these collections fall within 'type A'. The germination behaviour of seeds of collection P1 have been shown to differ from both of the previously mentioned groups. In analysis 7 it was noted that samples representing this collection were almost distinct from other samples of 'type A'. It was also pointed out that their position in the ordination could be considered intermediate between A. powellii 'type A' and A. retroflexus.

McWilliams et al (1968) have suggested that hybridisation between A. powellii and A. retroflexus may account for the occurrence of populations of A. retroflexus with seed dormancy. Hybridisation might also account for the population of A. powellii with abnormal germination behaviour observed here.

In the analyses that include the samples mentioned above together with the large number of other samples, the differentiation within A. powellii 'type A' is no longer visible. This can be interpreted in two ways. Either there is a continuum of variation of which only stages have been included in experimental work or else the differentiation is obscured by the introduction of environmentally induced variation in samples representing plants from diverse habitats. There is evidence to suggest that the latter explanation is correct, although this does not preclude the possibility of a continuum of variation. In those analyses that include all of the samples (analyses 2 or 3), samples that represented plants grown at the experimental farm

tended to segregate together within a small part of the total variation of  $\underline{A}$ . powellii 'type  $\underline{A}$ '.

Although the third reason for conducting this investigation was to establish whether there was evidence of hybridisation and introgression, the results of the analyses provide no answer to this problem, the plants described as putative hybrids occupied positions in the ordinations that supported However no light was shed on this view of their identity. Moreover, their high sterility makes it their ancestry. doubtful that recent and repeated hybridisations have been responsible for the patterns of variation observed. The sterility of these plants was such that it was often difficult and time consuming to obtain the required eight fruits in order to measure the characters that were used. Consequently, the choice of fruits could not be considered Some of the seeds that were to represent a random sample. measured were of doubtful viability (as determined from Thus it is perhaps not surprising that their appearance). the ordinations of these samples yield no clues as to their ancestry.

Table 3.25 presents the means and standard deviations of each character for the four taxa recognised.

THE MEANS AND STANDARD DEVIATIONS OF EACH TAXON, FOR THOSE CHARACTERS CONTRIBUTING MOST TO THE FACTOR LOADINGS

	oflexus	S.D.	0.07	90.0	0.11	20.0	0.11	0.18	0.34	0.05
	A. retroflexus	Mean	1.1%	0.79	2.00	4.98	0.84	0.74	5.29	7.
	allii B	S.D.	60.0	90.0	0.16	0.16	0.07	00.00	0.30	0.07
TAXON	A. powellii type B'	Mean	0.95	0.93	2.75	4.93	0.97	00.00	3.19	1.34
든		S.D.	0.11	0.07	0.15	0.35	0.17	0.04	0.43	0.07
	A. powellii type A'	Mean	1.17	0.73	2,92	4.39	6.47	0.02	2.73	1.30
	hybridus	S.D.*	90.0	90•0	0.16	0.18	0.09	0.01	0.25	0.05
	A. hybr	Mean	0.84	0.85	2.90	4.98	0.93	00.00	2.01	1.11
CHARACTER		Description	Distance from base of utricle to point of circumscission (mm)	Distance from point of circumscission to stylar sinus (mm)	Number of style branches	Number of tepals	Proportion of tepals with excurrent midribs	Proportion of tepals with retuse apices	Length of the longest tepal (mm)	Length of the seed (mm)
		No	~	2	2	, 4	7	Q	2	ω

(continued)

		A. hyb:	hybridus	A. powellii	91111 8 A'	A. powellii 'type B'	ellii e B'	A. retr	retroflexus
( <u>)</u>	Description	Mean	х. П.	Mean	S.D.	Mean	S.D.	Wean	S.D.
0	1000A								
0	Relative distance from micropyle to point of greatest seed width (mm)	0.52	0.02	0.58	0.03	0.57	00.0	0.55	0.02
10	Greatest width of the seed (mm)	1.03	0.03	1.07	0.05	1.09	0.04	1.02	0.04
25	Length of the bract (mm)	2.60	0.32	4.30	0.63	3.50	0.37	4.31	0•48
27	Ratio of the length of the utricle to the bract length	99•0	0.08	0.45	0.05	0.54	90.0	0.45	0.05
28	Ratio of the length of the longest tepal to the length of the bract	0.78	60.0	0.64	60.0	0.92	0.09	0.77	0.08
29	Length of terminal spike of the inflorescence (mm)	63.29	41.9	107.7	59.4	105.8	53.6	50.36	25.6
32	Width of the terminal spike at mid-length (mm)	9.36	1.82	15.13	2.92	14.26	12.04	17.12	2.78
35	Length of the third lateral branch (mm)	20•64	13.21	60,16	45.98	76.74	63.98	26.94	10.10
36	Width of the third lateral (mm)	6.07	1.21	11.69	3.29	9.90	2.83	12.68	2.90
								•	

CHARACTER

(continued)

_					
roflexus	S.D.	0.13	0.12	8.45	
A. reta	Mean	0.87	7-	19.62	
ellii e B'	S.D.	0.18	0.23	2.22	
A. power	Mean	0.75	1.28	5.17	
	S.D.	0.20	0.28	4.26	
A. pow	Mean	0.81	1.17	9.20	
	S.D.*	0.11	0.13	14.61	
A. hyb:	Mean	0.88	1.12	27.93	
	No. Description	39 Relative length of first lateral branch	40 Relative length of fifth lateral	41 No. of laterals in the 10 cm below the terminal spike	
	A. powellii A. powellii A. retroflexus 'type A' type B'	A. hybridus A. powellii A. powellii type A' type B' Description Wean S.D.* Mean S.D. Mean S.D.	Relative length of first 0.88 0.11 0.81 0.20 0.75 0.18	Relative length of fifth  Relative length of fifth  Relative length of fifth  1.12 0.13 1.17 0.28 1.28 0.25	Relative length of fifth   A. 10 cm   A. 1

\* Note: S.D. = standard deviation

#### CHAPTER 4

# GEOGRAPHICAL DISTRIBUTION OF THE SPECIES

### 4.1 Introduction

The sum of the environmental factors acting upon a species and the plasticity of its response are expressed in the geographical distribution of the species. Thus, potential insights into the ecology of the species may be gained from studying both the widespread and the local distributions.

# North American and global distribution

The large scale distribution of the three species included in this study has been reported in detail by McWilliams (1966). Sauer (1967) has given concise statements about the origins and distributions of these and other species in the section Amaranthus as follows:

"Amaranthus retroflexus L. is a native riverbank pioneer of the central and eastern U.S. and adjacent regions of southeastern Canada and northeastern Mexico. In much of this area it has become one of the commonest of all agricultural weeds... It has since become naturalized throughout temperate regions of the northern and southern hemispheres.

Amaranthus powellii is a pioneer of canyons, desert washes, and other open habitats, ranging through the western Cordilleran system of North and South America, with wide gaps in the wetter regions of Central America. The species has long been a common weed in the West.

It began appearing as a rare adventive in eastern North America about 1900; since 1940 it has become a widespread and troublesome weed there. The species also arrived in Germany as an adventive shortly before 1900 and has since become an abundant and still expanding weed of northern and central Europe ... It has also recently invaded southern India and South Africa.

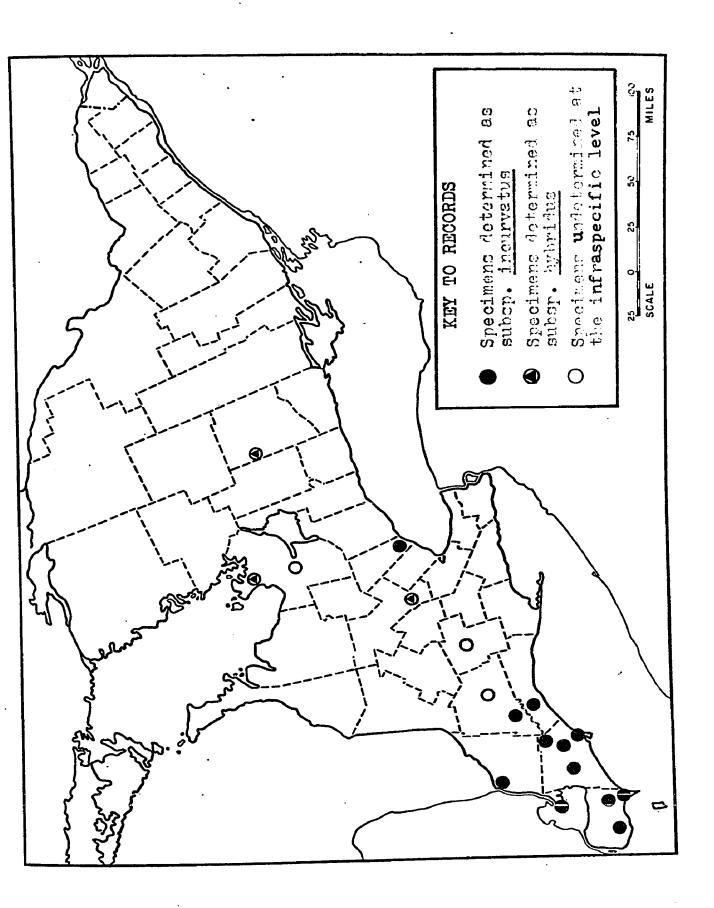
Amaranthus hybridus is evidently a native riverbank pioneer of milder and moister regions from eastern North America through Mexico and Central America to northernmost South America. It has long been a common field weed in this region. Its earliest and most successful emigration was to the Mediterranean region ... It has recently become a naturalized weed in western North America, eastern Asia, Australia, and South Africa."

# Distribution in southern Ontario

The local distribution of each species was assessed from information accompanying the specimens in several herbaria. Dr. C. Frankton kindly provided details of the specimens included in the herbarium of the Plant Research Institute, Canada Department of Agriculture, Ottawa (abbreviated DAO following the scheme published in "Index Herbariorum, see Lanjouw and Stafleu, 1954). Professor F.H.Montgomery gave valuable assistance in loaning indexed records of specimens in the major Ontario herbaria. Specimens were examined in the herbaria of the following institutions: Hamilton Botanic Gardens (HAM), McMaster University (MCM), National Museum of Canada (CAN), University of Guelph (OAC), University of Toronto (TRT) and University of Western Ontario (UWO).

The resulting distributions are presented in figures
4.1 to 4.3. Figure 4.1 consists of records for A. hybridus.

Fig. 4.1 The distribution of records of <u>Amaranthus</u>
<a href="https://doi.org/10.1001/journanthus">https://doi.org/10.1001/journanthus</a>
<a href="https://doi.org/10.1001/journanthus</a>
<a href="https://doi.org/10.1001/journanthus</a>
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The records for A.hybridus subsp. incurvatus indicate that it is restricted to the five extreme southwestern counties of the Province. The records for Elgin and Middlesex Counties have resulted from the present investigation. Only one specimen identified as subsp. incurvatus was recorded from outside the five southwestern counties. This plant was collected in the city of Toronto in 1904 and no information was provided concerning its habitat or abundance.

The known distribution of A. powellii in Southern

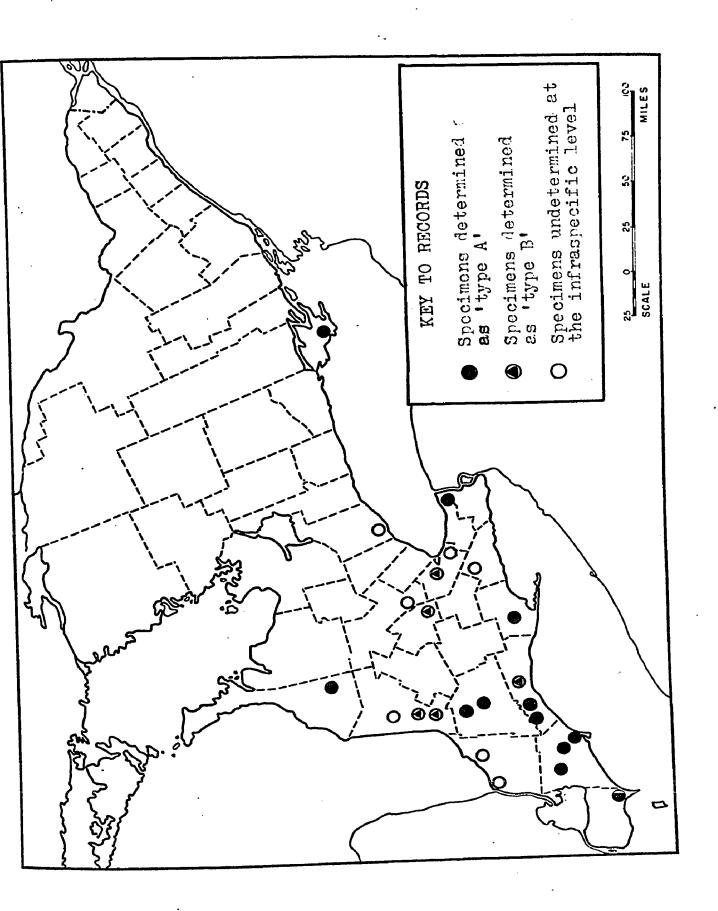
Ontario is recorded in figure 4.2. With the exception of material examined by Sauer or Frankton, most specimens of this species had been incorrectly identified as either

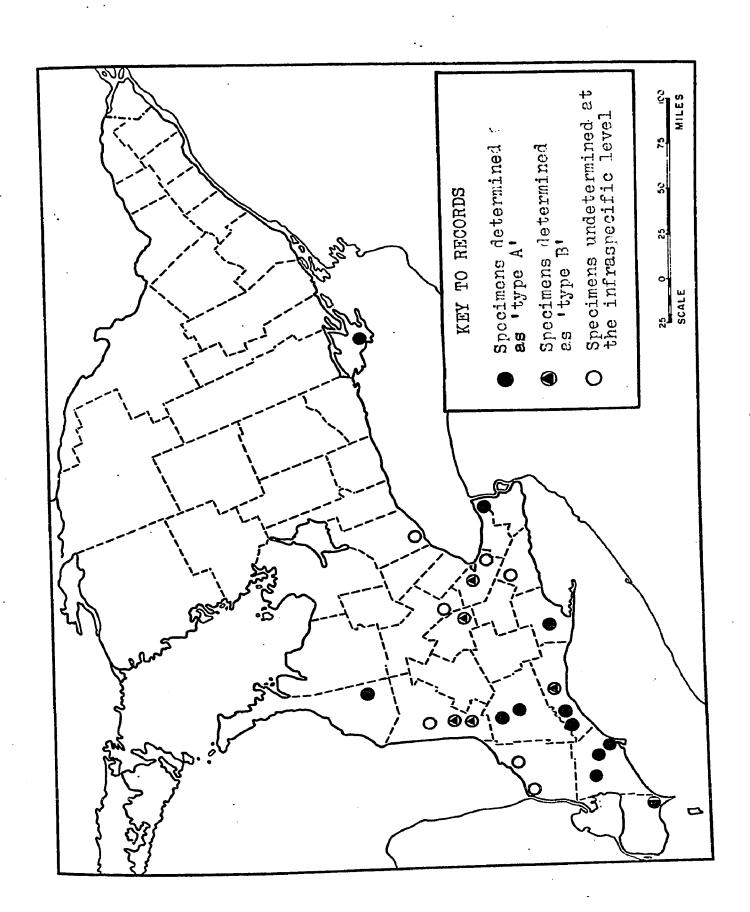
A. hybridus or A. retroflexus. Thus there is a possibility that some specimens of A. powellii exist among material that was not personally examined and thus are not recorded here.

In examining material at the Plant Research Institute,

Ottawa and at the University of Western Ontario it was

Fig. 4.2 The distribution of records of <u>Amaranthus</u>
<u>powellii</u> in Southern Ontario





possible to distinguish between specimens of the two 'types' discussed on pages 97 and 98. These distinctions are included in figure 4.2.

The evidence suggests that A. powellii is also restricted in its distribution in Southern Ontario. It has a wider distribution than A. hybridus subsp. incurvatus but is absent from the eastern, and central parts of the region. There are no obvious differences in the distribution of the two 'types' of A. powellii.

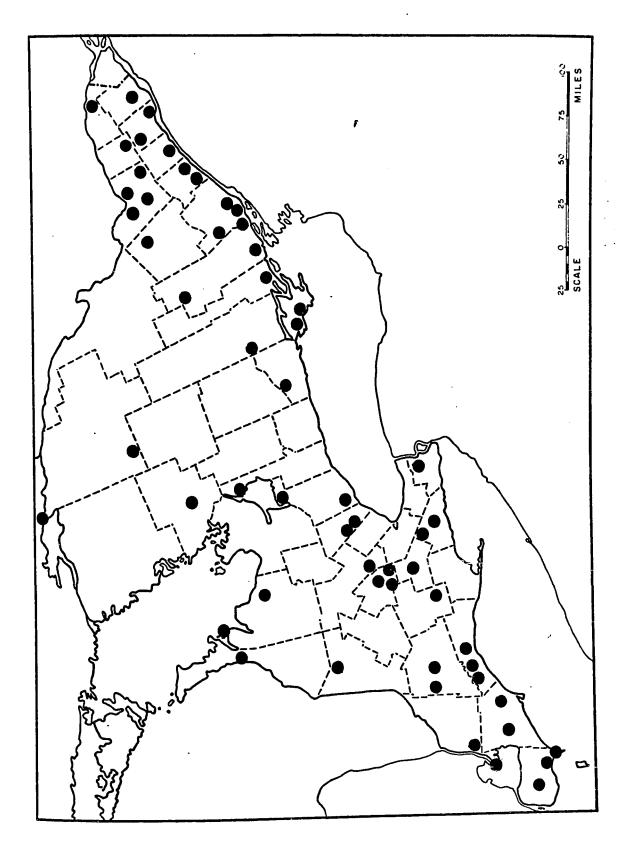
Figure 4.3 presents the recorded distribution of

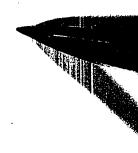
A. retroflexus, which is based on all records from the sources
mentioned. The species extends also into Northern Ontario,
to the Manitoba border and beyond. Discontinuities in
distribution within Southern Ontario probably reflect
sampling deficiencies, since most records come from the
immediate vicinity of herbaria or agricultural research
stations. The widespread distribution of records suggest
that the species occurs throughout Southern Ontario.

4.2 Surveys of the local distribution4.2.1 Purpose

Two surveys of the distribution of the three species in the local area were planned in an attempt to provide answers to the following questions: (1) How frequently do each of the species occur? (2) Is any species restricted to, or more frequently found in a particular habitat? (3) Are any of the species capable of exploiting a common habitat

Fig. 4.3 The distribution of records of <u>Amaranthus</u>
retroflexus in Southern Ontario





and how frequently do they do so? (4) Is the frequency of any species affected by agricultural practices, types of crops or edaphic conditions?

### 4.2.2 Methods

In the first survey several parameters were recorded for a limited number of sites that were chosen irrespective of the presence of <u>Amaranthus</u>. In the second survey a less rigorous sampling technique was employed in order to include many more sites.

### Survey 1

### a) Area of study

An area was chosen to include (a) known localities of each of the three species and (b) different soil types representative of the range encountered in southwestern Ontario. For these reasons and for convenience the land area described by Canada Department of National Defence Map 40 1/12 (Bothwell East, scale 1:50,000) was chosen for sampling. The area was approximately 20 km East to West and 26 km North to South, and included parts of the counties of Elgin and Middlesex. On the southeast side the area was bounded by Lake Erie.

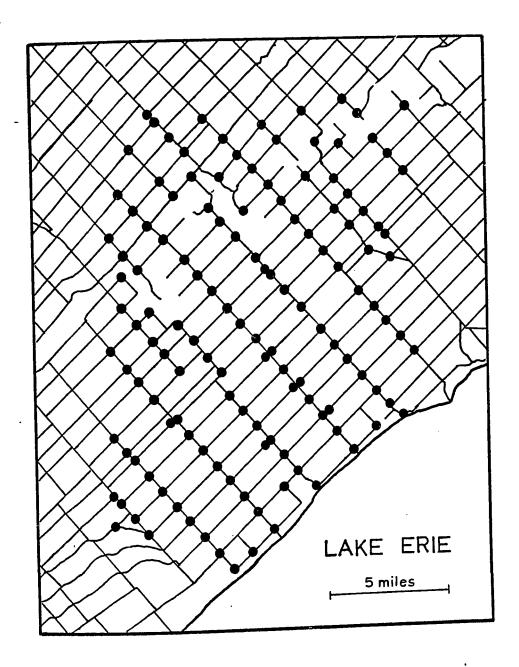
# b) Sampling plan

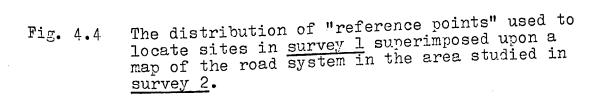
The decision was made to sample 100 sites within the area of study. The sites were determined in the following way. Random numbers were employed to obtain 100 points described by coordinates on the 1000 metre Universal Mercator Grid that was superimposed on the map used. The nearest

physiognomic or cultural feature (shown on the map) to each of these points was designated as a "reference point" if it appeared that this reference point would be easily recognised from the road. Random numbers also were used to predetermine a direction that would be taken from each reference point. The remaining criteria were applied in the field.

The important role played by roads in conducting this survey was justified by their regular and orthogonal arrangement. In fact, most of the reference points that were described were intersections of perpendicular roadways (Fig. 4.4) and the directions to be taken from a reference point were restricted to directions in which travel was possible by car.

Species of Amaranthus have neither been reported nor observed as weeds of pasture land, and thus a first criterion was to include only arable fields as sites. A second criterion, this time of convenience, was to include only fields adjacent to the road or one field away from it. The most important criterion that had to be satisfied before a field was included was that information concerning the cultural history of the crop could be obtained. Thus, while travelling from a feference point in a predetermined direction, the field that was sampled was the first field encountered on the right side of the road that satisfied the criteria listed above.





In some instances no field was encountered that satisfied the criteria for sampling before an adjacent reference point was reached. When this happened the journey was repeated with consideration given to fields on the left side of the road. If this approach was unsuccessful a new direction was taken from the reference point and the procedures were repeated. Occasionally no site could be found that met the requirements and the sample was abandoned. This happened primarily in areas with little arable land and reduced the total sample number.

When a field had been selected, sampling was restricted within a 20 m by 20 m area in the corner first encountered.

## c) Parameters measured

The first information sought at each site concerned the cultural history of the field. The following details were recorded:

- 1) The kind of crop present,
- 2) The crop grown in that field in the previous season,
- 3) The time at which the soil had been ploughed following the previous crop,
- 4) The use of fertilizers and synthetic herbicides with the present crop.

The sample area was then visited and the presence or absence of each of the three species of Amaranthus was determined. A soil sample was taken at a point ten metres from the corner of the field in the direction away from the road and four metres from the side of the field. This

sample was taken back to the laboratory for analysis. Finally the position of the sample was recorded to the nearest 100 m using the Universal Mercator Grid on the Department of National Defence map.

# d) Soil Analysis

The purpose in collecting and analysing soil samples was to determine whether the soil in the sample site did in fact belong to the soil type described in Soil Survey Reports numbers 2 and 6 (Canada Department of Agriculture). For this reason, an analysis of the texural constitution of each sample was made. The method of textural analysis that was employed followed procedures used in the Soil Mechanics Laboratory of the Faculty of Engineering, University of Western Ontario. These procedures were basically identical to those described by Piper (1944). A description of the procedural details is included in Appendix 4.

# Survey 2

# a) Area of study

The area of survey 2 included the area described for survey 1 together with an additional 4 km border surrounding the area on the north, west and parts of the south and east sides.

# b) Sampling plan

Two hundred and seventy seven sites were sampled on the basis of the network of roads that formed an orthogonal grid throughout the area (see Fig. 4.4). A site was defined as any point between two reference points on the map that

delimited the sample. Within these limits the first field of each crop type was sampled if it was observed to contain any one of the species.

### c) Parameters measured

For each sample, the crop type, species of Amaranthus present, and the grid reference were recorded.

### 4.2.3 Results

The information recorded in surveys 1 and 2 is presented in Appendix 2, in tables A2.1 and A2.2 respectively. The results of the analyses of soil samples are presented in Appendix 2, table A2.5.

4.2.4 Analysis of the results

### 4.2.4.1 Methods of analysis

Statistical techniques were employed to determine the probable correctness of answers given to questions raised on page 110. Each question was defined in terms of a statistical hypothesis that could be tested by using the data collected in the surveys.

The data were easily displayed in contingency tables of two or three dimensions. These tables were investigated using the methods of information analysis described by Kullback (1959) and Kullback, Kupperman and Ku (1962). Kullback et al (1962) compare the uses of  $\chi^2$  and their "minimum discrimination information statistic" and conclude:

"The utility of the m.d.i.s., however, lies in its additivity, convexity, and computational properties.
. . . the m.d.i.s. can be analysed into several additive components, similar to the analysis of additive components, similar to the analysis of variance, for a hypothesis that is equivalent to the variance, for a hypotheses that are of interest. combination of several hypotheses that are of interest. Each component of the m.d.i.s. is itself an m.d.i.s. Each component of the m.d.i.s. is itself an m.d.i.s.

degrees of freedom".

The computational properties mentioned above refer to the ease with which the statistic can be evaluated. If tables of  $\underline{n}$  ln  $\underline{n}$  are available, computation is restricted to reading values from the tables and performing additions and subtractions.

The m.d.i.s. was chosen for use in this analysis because of the ease with which the information content of contingency tables can be analysed into additive components by using this method.

### Analysis of 2-way tables

Where the data can be classified according to two different criteria, a null hypothesis that the two criteria are independent can be tested using the following statistic (Kullback et al, 1962):

$$2I = \sum_{i=1}^{r} \sum_{j=1}^{c} 2f_{ij} \ln f_{ij} + 2n \ln n - \sum_{i=1}^{r} 2f_{i} \cdot \ln f_{i}$$
$$- \sum_{j=1}^{c} 2f_{i} \cdot \ln f_{i}$$

where:

fij = the frequency in a particular cell in the
 table

$$f_{i.} = \sum_{j=1}^{c} f_{ij}$$
 (row totals)

$$f_{ij} = \sum_{i=1}^{n} f_{ij}$$
 (column totals)

$$n = \sum_{i=1}^{c} \sum_{j=1}^{c} f_{ij}$$

r = the number of rows

c = the number of columns

The m.d.i.s. (2I) is asymptotically distributed as  $\chi^2$  with (r-1)(c-1) degrees of freedom under the hypothesis that

the row and column classifications are independent.

Analysis of three-way tables

Where the data can be classified according to three different criteria (designated as rows, columns and depth), there are three hypotheses of particular interest. These are the hypotheses of first order independence between the first and second, the second and third, and the first and third classifications. Kullback (1959: chapter 8, section 3.5) has described the equations that are used to determine the components of information that are required to test these hypotheses. The analysis can be set out in a table that resembles the table for the analysis of variance:

### Component due to

## <u>Information</u>

egrees of freedom

1) Rows x columns 
$$2\sum_{i=1}^{r}\sum_{j=1}^{c}x_{ij}$$
.  $\log \frac{n x_{ij}}{x_{i}...x_{ij}}$  (r-1)(c-1)

2) Columns x depth 
$$2\sum_{j=1}^{c}\sum_{k=1}^{d}x_{\cdot jk}\log\frac{n x_{\cdot jk}}{x_{\cdot j} \cdot x_{\cdot \cdot k}}$$
 (c-1)(d-1)

3) Rows x depth 
$$2\sum_{i=1}^{r}\sum_{k=7}^{d}x_{i\cdot k}\log\frac{n x_{i\cdot k}}{x_{i\cdot \cdot \cdot}} \qquad (r-1)(d-1)$$

The information belonging to the first component can be computed from the following equation:

ated from the following equation:
$$2I = n \ln n + \sum_{i=1}^{c} \sum_{j=1}^{c} x_{ij} \cdot \ln x_{ij} \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \ln x_{i} \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \ln x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot \cdot - \sum_{i=1}^{c} x_{i} \cdot \cdot \cdot -$$

The information belonging to the second and third components can be computed from similar equations that differ

only in subscripts. The value for 2I for each component is asymptotically distributed as  $\chi^2$  with appropriate degrees of freedom under each hypothesis of independence between classifications.

4.2.4.2 Hypotheses tested among the data from survey 1

## i) That each species was distributed with the same frequency

TABLE 4.1

THE FREQUENCY OF THE SPECIES IN 89 CULTIVATED FIELDS

	Species	No. of sites occupied	Percentage frequency
Α.	hybridus	11	12.4
Α.	powellii	50	56.3
Α.	retroflexus	69	77•5

The frequency of the three species in survey 1 are presented in table 4.1. Three 2-way tables were prepared in which sites were classified for each of two species into sites with the species and sites without the species. In each table the hypothesis was tested that the classification for one species was independent of the classification for the other species. Values computed for the m.d.i.s. (2I) for each pair of species are presented in table 4.2.

The results indicate that for each pair of species the hypothesis should be rejected. Thus the frequencies with which each species was present in survey 1 were significantly different.

#### TABLE 4.2

# ESTIMATES OF INDEPENDENCE (21) BETWEEN THE FREQUENCIES OF OCCURRENCE OF PAIRS OF SPECIES

A. hybridus A. powellii

A. powellii

40.25\*\*

A. retroflexus

83.25\*\*

9.26\*\*

Note: \*\* - The probability that these estimates represent chance departures from the null hypothesis is less than 0.01.

# ii) That the presence of one species was independent of the presence of one of the other species

The data were arranged in a three-way contingency table in which classifications were made on the basis of the presence or absence of each species from a site (table 4.3). Information in the contingency table was analysed into three components that permitted hypotheses of independence between each pair of species to be tested. Table 4.4 presents the analysis of information.

#### TABLE 4.3

# A CONTINGENCY TABLE OF THE OCCURRENCE OF EACH SPECIES

A. hybridus

Present Absent

A. retroflexus Present Absent Present Absent

Present 9 1 35 24

Absent 0 1 6 13

TABLE 4.4

THE ANALYSIS OF INFORMATION TO DETERMINE ESTIMATES OF INDEPENDENCE BETWEEN SPECIES

Component due to	2I	Degrees of freedom	Significance '
H x R <sup>2</sup>	1.53	1	ns
R x P	7.23	1	* *
нхР	3.66	1	ns

Notes: 1 - The probability that the value of 2I represents a random departure from independence is:

ns - greater than 0.05
\*\* - less than 0.01

2 - Species are abbreviated as follows:

H = A. hybridus P = A. powellii

R = A. retroflexus

The results indicate that the hypothesis should be accepted for A. hybridus and A. powellii, and A. hybridus and A. retroflexus, and rejected for A. powellii and A. retroflexus. Rejection of the hypothesis for A. powellii and A. retroflexus signifies that these species were correlated in their occurrence in the sense that the two species occurred together more frequently than would be predicted on the basis of random expectations.

# iii) That the occurrence of any species was independent of the addition of fertilizer

The data were arranged in three 2-way tables (Table 4.5) in order to test the hypotheses that the occurrence of each species was independent of the addition of fertilizer. Table 4.6 presents the values of the m.d.i.s. obtained in each table.

TABLE 4.5

CONTINGENCY TABLES FOR THE PRESENCE OR ABSENCE OF EACH SPECIES AND THE ADDITION OR NON-ADDITION OF FERTILIZER

Species and occ	urrence	With fertilizer	Without fertilizer
A. hybridus	present	10 71	1 7
A. powellii	present	49 32	5 3
A. retroflexus	present absent	64 17	8 0

TABLE 4.6

# VALUES FOR 2I FOR THE OCCURRENCE OF EACH SPECIES CLASSIFIED ACCORDING TO FERTILIZER USE

#### SPECIES

	A. hybridus	A. powellii	A. retroflexus
2I	0.00	0.01	2.58
<u> </u>	<b>-</b> • • • • • • • • • • • • • • • • • • •		ns
Significance	ns	ns	

Note: ns = The probability that the value of 2I represents a random departure from independence is greater than 0.05.

The results indicate that the hypothesis should be accepted for each species. In other words, the frequency of occurrence of each species was independent of the addition of fertilizer.

# iv) That the occurrence of each species was independent of the time of ploughing and the addition of synthetic herbicides

The data were arranged in three 3-way tables in order to test the hypothesis that the distribution of each species was independent of the time at which the field had been ploughed and the addition of synthetic herbicides.

Contingency tables for each species, classified according to these factors, are presented in table 4.7. Table 4.8 presents the results of the analysis of information.

TABLE 4.7

CONTINGENCY TABLES OF THE OCCURRENCE OF
EACH SPECIES CLASSIFIED ACCORDING TO
TIME OF PLOUGHING AND USE OF HERBICIDE

				Spring-ploughed		
		Autumn-I	ploughed	philip brond		
	pecies and ccurrence	With herbicide	Without herbicide	With herbicide	Without herbicide	
	hybridus				7	
	present	2	2	3	3	
	absent	10	19	32 	13 	
Α.	powellii			40	10	
	present	7	14	17		
	absent	5	7	18 	6	
Α.	retroflexus			Οli	13	
	present	8	20	24	•	,
	absent	4	1	11	<u> </u>	

TABLE 4.8

THE ANALYSIS OF INFORMATION TO DETERMINE ESTIMATES OF INDEPENDENCE BETWEEN OCCURRENCE, TIME OF PLOUGHING AND USE OF HERBICIDES FOR EACH SPECIES

Component due to	Species	2I	D.F. <sup>1</sup>	Significance <sup>2</sup>
Occurrence x ploughing	A. hybridus	0.00	1	ns
	A. powellii	0.94	1	ns
	A. retroflexus	2.91	1	ns
Occurrence x	A. hybridus	0.15	1	ns
herbicide	A. powellii	1.62	1	ns
	A. retroflexus	4.50	1	*
Ploughing x herbicide		8.54	1	**

Notes: 1 - D.F. = degrees of freedom

2 - The probability that the value of 2I represents a random departure from independence is:

ns - greater than 0.05

\* - less than 0.05

\*\* - less than 0.01

In the light of the results, the hypothesis that occurrence was independent of the time of ploughing should be accepted for each species. The hypothesis that occurrence was independent of the addition of synthetic herbicides should be accepted for A. hybridus and A. powellii but rejected for A. retroflexus. The presence of A. retroflexus was negatively associated with the addition of synthetic herbicides.

In this analysis a further hypothesis was tested; the hypothesis that addition of herbicide was independent of

the time of ploughing. The results indicate that this hypothesis should be rejected. Thus the results of this survey demonstrate a positive association between the addition of herbicides and spring ploughing.

### v) That the occurrence of each species was independent of the present or previous crop

The data were arranged in three 3-way tables in order to test the hypotheses that the occurrence of each species was independent of the nature of the present or previous crop (Table 4.9). The results of the analysis of information are presented in table 4.10.

TABLE 4.9 CONTINGENCY TABLES FOR THE OCCURRENCE OF EACH SPECIES CLASSIFIED ACCORDING TO PRESENT AND PREVIOUS CROP

	Provious Present crop						
Species and occurrence	Previous crop	Tobacco	Beans	Corn	Others*		
OCCUTTCHCC							
A. hybridus							
present	Tobacco Beans Corn Others*	0 0 1 2	0 3 1 0	0 1 2 1	0 0 0		
absent	Tobacco Beans Corn Others*	4 0 2 8	0 10 0 6	1 5 32 7	0 0 0 1		
A. powellii		<b></b>	0	1	0		
present	Tobacco Beans Corn Others*	2 0 3 4	0 10 1 3	4 16 5	0 0 1		
absent	Tobacco Beans Corn Others*	2006	0 3 0 3	0 2 18 3	0 0 0		
*** *** *** *** *** *** *** *** *** *** ***	O 01161 p		مين دين مين مين مين مين مين مين مين <u>مين مين مين مين مين مين مين مين مين مين </u>	(cc	ntinued)		

Species and occurrence	Previous crop	Tobacco	Present Beans	crop Corn	Others*
A. retroflexus					
present	Tobacco Beans Corn Others*	4 0 2 1	0 12 1 0	1 6 20 14	0 0 0
absent	Tobacco Beans Corn Others*	0 0 1 1	0 1 0 0	0 0 14 3	0 0 0 0

\*Note: Crops included as "others" were: cabbage, cucumber, hay, oats, pasture, tomatoes and wheat.

TABLE 4.10

INFORMATION ANALYSIS OF ESTIMATES OF INDEPENDENCE BETWEEN PRESENT CROP, PREVIOUS CROP AND THE OCCURRENCE OF EACH SPECIES

Component due to	Species	2I	D.F. <sup>1</sup>	Significance <sup>2</sup>
Occurrence x	A. hybridus	2.57	3	ns
previous crop	A. powellii	2.84	3	ns
	A. retroflexus	12.61	3	**
Occurrence x	A. hybridus	2.47	3	ns
present crop	A. powellii	2.77	3	ns
	A. retroflexus	10.29	3	**
Previous crop x		60.62	9	* *
present crop				

Notes: 1 - D.F. = degrees of freedom

2 - The probability that the value of 2I represents a random departure from independence is:

ns - greater than 0.05
\*\* - less than 0.01

Interpretation of the results of this analysis is not entirely straightforward. Each contingency table (Table 4.9) included a large number of classes and consequently the frequencies in many of these classes were zeros. In this situation the results of the analysis are not reliable and acceptance or rejection of hypotheses should be considered tentative.

The results indicate that the hypothesis of independence between present crop and the occurrence of a species should be accepted for A. hybridus and A. powellii but rejected for A. retroflexus. Likewise, the hypothesis of independence between previous crop and the occurrence of a species should be accepted for A. hybridus and A. powellii but rejected for A. retroflexus. The interactions between the occurrence of A. retroflexus and each of these classifications reflect positive associations between the presence of A. retroflexus and corn as the present and previous crop.

The results of this analysis also indicate a positive association between present crop and previous crop.

# vi) That the occurrence of each species was independent of the characteristics of the soil

The results of the textural analysis of soil collected at each site are presented in Appendix 2, table A2.5. Each site was described by the five characters evaluated in the textural analysis. These five characters were the weights of soil in each of five conventional particle-size classes expressed as percentages of the total weight of the soil sample. In order to test the hypothesis that the occurrence

of each species was independent of the characteristics of the soil, it was necessary to assign the sites to classes on the basis of their soil characters. With each site described by these five parameters it was difficult to visualise the relationship between sites. Consequently principal components analysis was used to simplify the description of variation among the sites. The technique of this analysis is described in chapter 3 on pages 38 to

. Its main effect is to reduce the number of dimensions in which the variation among the samples is described.

In order to perform a principal components analysis it is necessary to compute a matrix of similarity values between either characters or samples. Since the number of characters was less than the number of samples a matrix of similarities between characters was appropriate. of the characters was measured in the same units, (percent by weight) there were no strong reasons for choosing between the different coefficients of similarity that are available. (See Chapter 3 page 39). The dispersion coefficient was chosen for use in this analysis and a dispersion matrix (matrix of sums of squares and products) was computed. AnR-type principal components analysis was performed upon this matrix and five component axes were extracted. The eigenvalues for the first and second axes were 603.0 and 223.0 Together these two axes accounted for 98% respectively. of the variation in the data.

The characters that contributed the greatest amounts of variation to the first and second components were respectively the percentage of fine sand and the percentage of coarse sand. The values for the contributions of each character to these two components are given in table 4.11.

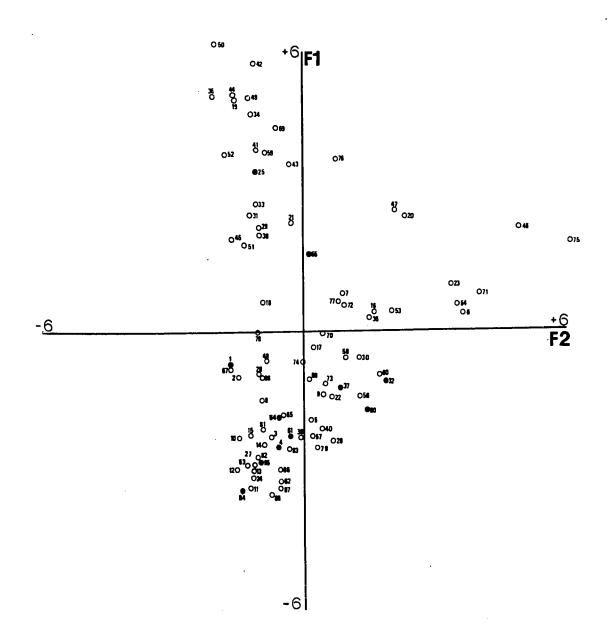
TABLE 4.11

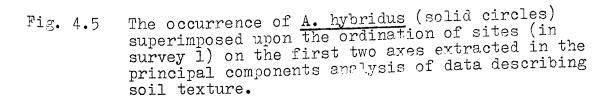
THE CONTRIBUTIONS OF VARIATION BY EACH CHARACTER TO THE FIRST TWO COMPONENT AXES (THE VALUES ARE ADJUSTED SO THAT THEY SUM TO UNITY FOR EACH COMPONENT)

#### Characters

	1	2	3	4	5
Component	% gravel	% coarse sand	% fine sand	% silt	% clay
	002	037	+.825		
2	+.007	+.855	263	400	200

The ordination of sites on the first two component axes is present in figure 4.5. The ordination reveals a continuum of variation with the indication of clusters at both extremes of the first axis. Each site has been identified, in the ordination, by number. In figure 4.8 sites have been identified by soil type as determined from Soil Survey Reports number 2 and 6 (Canada Department of Agriculture). Some sites appear to have been misdetermined on the basis of the Soil Survey Reports; for example sites 25, 31 and 59. However, the majority of sites show agreement between their determination from the Soil Survey





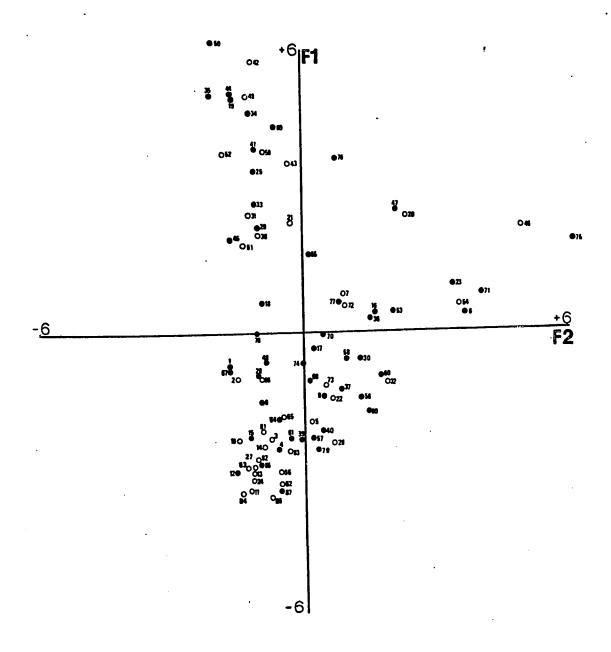


Fig. 4.6 The occurrence of A. powellii (solid circles) superimposed upon the ordination of sites (in survey 1) on the first two axes extracted in the principal components analysis of data describing soil texture.

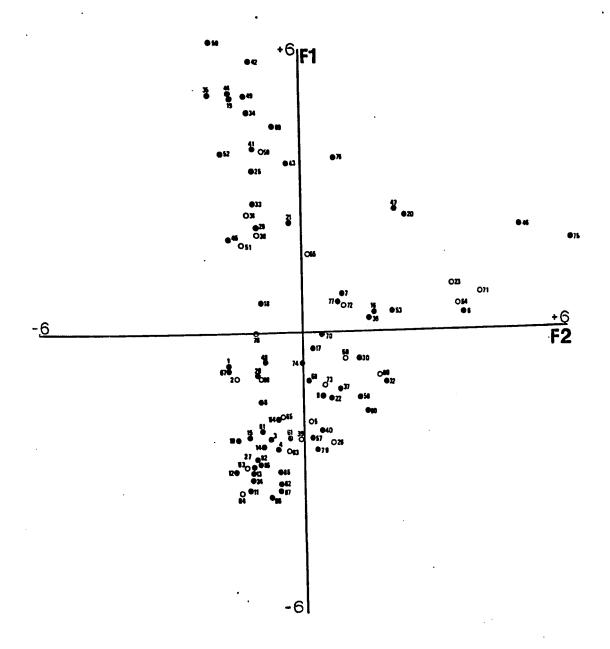
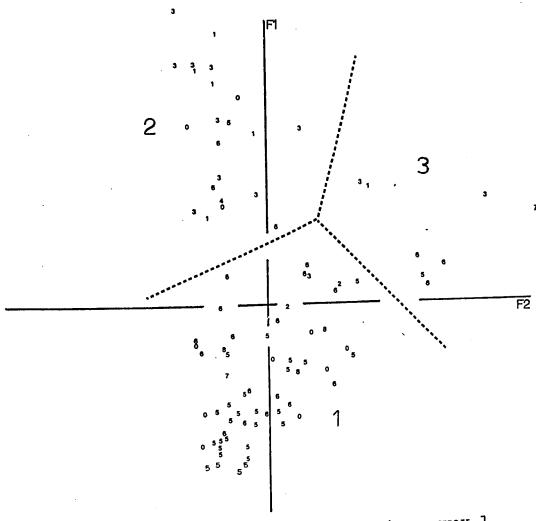
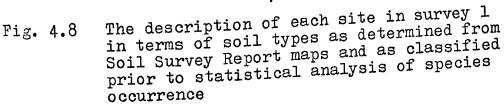


Fig. 4.7 The occurrence of A. retroflexus (solid circles) superimposed upon the ordination of sites (in survey 1) on the first two axes extracted in the principal components chalysis of data describing soil texture.





The large numbers (1 to 3) refer to the soil classes described for the purpose of statistical analysis.

The small numbers indicate the position of each site on the ordination of soil data and describe the soil type as determined from Soil Survey Report maps as follows: 2 - Clyde clay loam

1 - Brookston clay loam

4 - Perth clay

6 - Brookston sandy loam 3 - Haldimand clay

5 - Berrien loamy sand 7 - Brady gravelly loam 8 - Fox gravelly loam

0 - Indeterminable, see table A2.1, note 4

Reports and their position on the ordination. The two clusters identified in the ordination correspond to sandy loams and clays, although there is considerable variation within each cluster. Figures 4.5 to 4.7 portray the occurrence of each species superimposed upon the ordination.

The ordination of sites on two axes which accounted for 98% of the variation simplified the task of assigning the sites to classes. Classes were defined arbitrarily. The temptation to describe a large number of small and tightly clustered groups was weighed against the advantage of describing a small number of groups; a greater sensitivity of statistical tests based on large withingroup numbers and small inter-group degrees of freedom. The advantages of a small number of groups were preferred and three classes were described as shown in figure 4.8.

Three 2-way contingency tables were prepared in which the sites were classified according to the soil classes described above and the presence or absence of each species. These contingency tables are presented in table 4.12.

TABLE 4.12

CONTINGENCY TABLES FOR THE OCCURRENCE OF EACH SPECIES CLASSIFIED ACCORDING TO SOIL CLASSES

Species and o	ccurrence	1	Soil class 2	3
	present	0	2	9
A. hybridus	absent	8	20	49
			(continued)	

Species and occurrence		1	Soil class 2	3
A. powellii	present	4	13	37
	absent	4	9	21
A. retroflexus	present absent	6 2	18 4	43 15

The contingency tables were analysed in order to test the hypothesis that the occurrence of each species was independent of soil characteristics. Values obtained for the m.d.i.s. (2I) were 3.44 for A. hybridus, 1.22 for A. powellii and 1.15 for A. retroflexus. None of these values were significant at p = 0.05 suggesting that the hypothesis should be accepted for each species. Thus the occurrence of each species was independent of the class of soil when the classes were described in the manner discussed. 4.2.4.3 Hypotheses tested among the data of survey 2

Observations were made only at sites in which at least one of the three species of Amaranthus was present. No measure was made of the frequency of such sites compared with sites containing none of the species, and this fact makes interpretation of the results difficult. However, if each species was equally easy to detect, then the relative frequencies of the species as determined by the two surveys should have been the same. The hypothesis that the relative frequencies of the three species were independent of the survey was tested statistically in the

following manner. The data from surveys 1 and 2 were arranged in a contingency table, excluding data from survey 1 for sites with none of the species (Table 4.13). The results of the analysis of information are presented in table 4.14.

TABLE 4.13

CONTINGENCY TABLE OF THE PRESENCE AND ABSENCE OF EACH SPECIES IN SITES WITH AT LEAST ONE SPECIES IN SURVEYS 1 AND 2

	A. hybridus present absent		A. powellii present absent		A. retroflexus present absent	
Survey 1	11	65	50	36	69	7
Survey 2	55	219	201	73	258	16

**TABLE 4.14** 

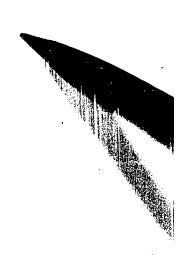
INFORMATION ANALYSIS OF ESTIMATES OF INDEPENDENCE BETWEEN SPECIES, OCCURRENCE AND SURVEYS

Component due to	2I	Degrees of freedom	Significance
Species x occurrence	222.6	2	**
Occurrence x survey	1.1	1	ns

Note: 1 - The probability that the value of 2I represents a random departure from independence is:

ns - greater than 0.05
\*\* - less than 0.01

The results indicate that the hypothesis of independence between relative frequencies of the species and the survey in which they were determined should be accepted. In other words, the relative frequencies of the three species did not



differ significantly between surveys. The highly significant component of interaction between species and occurrence indicates that in the two surveys there are differences in the frequencies of individual species.

4.2.5 Discussion

4.2.5.1 Discussion of the relationships among the data from survey 1

### a) Differences in frequency

Under uniform conditions that were probably extremely favourable for growth, plants of each species were seen to produce approximately the same quantities of seeds (during germination experiment 9). Therefore the observation that the three species occur with different frequencies in the field suggests that environmental factors act differentially to limit the biotic potential of each species.

# b) Association between the occurrence of A. powellii and A. retroflexus

The positive association between the occurrence of these two species indicates that they resemble each other more closely in their responses to the environment than either of them resembles A. hybridus. However this interpretation must be qualified by the fact that the number of sites that included A. hybridus was small whereas the number of sites including A. powellii, and the number including A. retroflexus were both large (Table 4.1). At the same time this observation indicates that there were more sites in which either both species (A. powellii and A. retroflexus) were present or both species were absent than would be predicted from random expectations.

# c) Association between the occurrence of A. retroflexus and the addition of herbicide

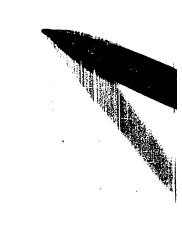
Two reasons can be suggested for the observation that A. retroflexus occurred less frequently in fields receiving an application of a herbicide, while the occurrence of the other two species was independent of this factor. A. retroflexus was seen to be the most frequent species from which it can be assumed that the species is adapted to a greater number of different environments than A. hybridus Consequently if each species were equally and A. powellii. susceptible to herbicides but  $\underline{A.\ hybridus}$  and  $\underline{A.\ powellii}$ were greatly restricted by other factors then their relationship to herbicide would be less pronounced than that of Secondly, A. retroflexus may be more A. retroflexus. susceptible to herbicides than A. powellii and A. hybridus. Gleason and Cronquist (1963) described the vegetative parts of A. retroflexus as "finely villous especially upward", and Thus the leaf and those of A. powellii as subglabrous. stem surfaces of A. retroflexus would probably retain droplets of foliar contact herbicides longer than those of Plants of A. hybridus observed in the present A. powellii. investigation had an indumentum resembling that of A. retroflexus which would suggest that their retention of Differences may also herbicide droplets would be similar. exist between the species in the response of their seeds and seedlings to pre-emergence herbicides.

# 4) Association between the occurrence of A. retroflexus and the type of crop

The relationship between the occurrence of A. retroflexus and the present and previous type of crop can probably be explained in part by the association between crops and the application of herbicides. A. retroflexus occurs at a lower frequency in corn fields than in other crops. A herbicide that is frequently used in corn is atrazine which may retain activity in the soil for more than 12 months (Talbert and Fletchall, 1964). It was observed that the present crop was not independent of the previous crop in the sites sampled. This meant that for a field containing corn at the time of the survey there was a high probability that corn had been the crop in the previous season. The persistence of atrazine may explain the absence of A. retroflexus from corn fields which had not been treated with herbicide in the year that the survey was made. In these sites the persistence of atrazine from an application in the previous year may have been sufficient to eliminate A. retroflexus.

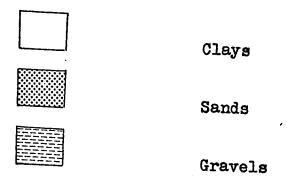
# 5) Independence between species occurrence and characteristics of the soil

Defore commenting on the relationship between species occurrence and characteristics of the soil there is further information to be presented. In figures 4.9 to 4.11 the occurrence of each species is indicated on maps which identify each site. There is a transparent overlay with each map that records the major soil differences within the area of study, as determined from Soil Survey Reports. Soil



## Figs. 4.10 to 4.15

Transparent overlays to these figures illustrate the distribution of major areas of different soils classified according to table 4.14. The symbols used in these overlays are as follows:



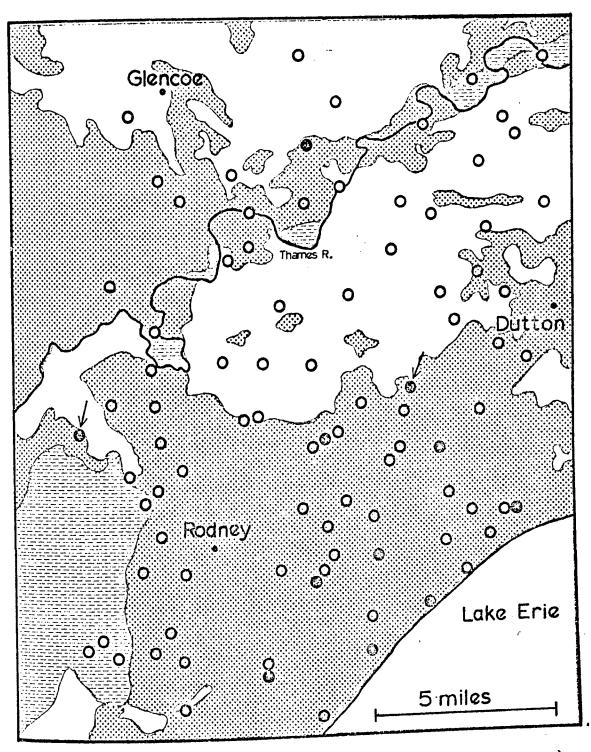


Fig. 4.9 The occurrence of A. hybridus (solid circles) superimposed upon the distribution of sites in survey 1. Sites 25 and 55 are marked with arrows and are discussed in the text. (See facing page for key to the overlay.)

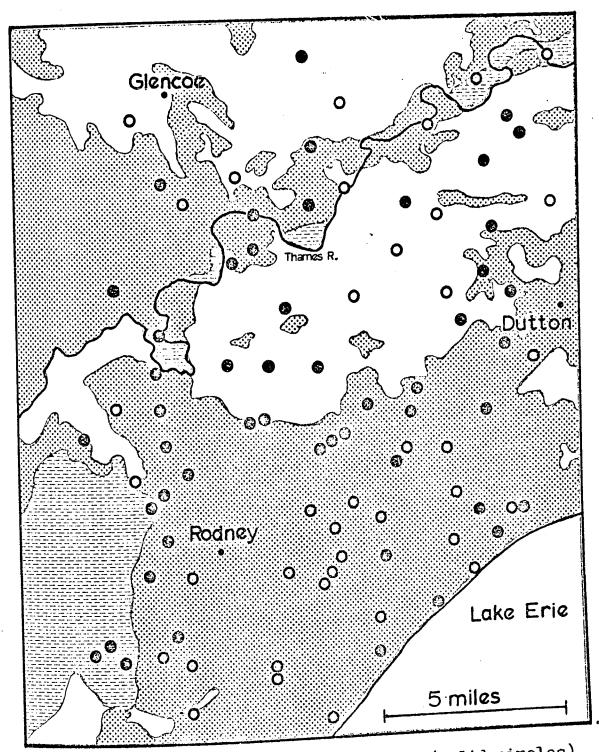


Fig. 4.10 The occurrence of A. powellii (solid circles) superimposed upon the distribution of sites in survey 1. (See the page facing Fig. 4.9 for a key to the overlay.)

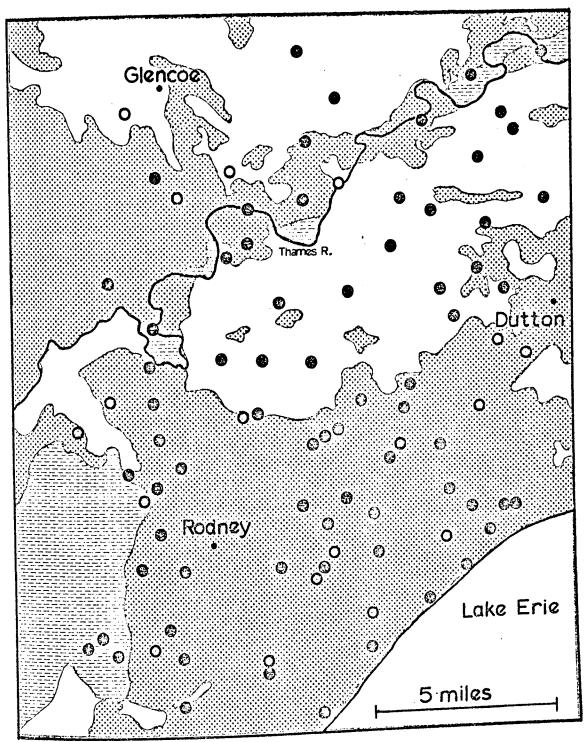


Fig. 4.11 The occurrence of A. retroflexus (solid circles) superimposed upon the distribution of sites in survey 1. (See the page facing Fig. 4.9 for a key to the overlay.)

types described in the Soil Survey Reports have been combined and illustrated as clays, sands and gravels (Table 4.15).

#### **TABLE 4.15**

THE COMBINATION OF SOIL TYPES INTO THE MAJOR GROUPS THAT ARE INCLUDED IN FIGURES 4.9 TO 4.14

Major group	Soil types (from Soil Survey Reports) included in major group
Clays	Brookston clay loam Clyde clay Haldimand clay Perth clay
Sands	Berrien loamy sand Berrien sand Brookston sandy loam Granby sand Mixed, mainly Ottawa sand and Miami silty clay loam
Gravels	Brady gravelly loam Fox gravelly loam

A. hybridus were present were all distributed within or adjacent to areas of sands. This suggests a greater association between the occurrence of this species and characteristics of the soil than was indicated from the analysis of soil samples from each site. In the ordination of sites in terms of their particle size distribution, (Fig. 4.5) two sites containing A. hybridus occupied positions among sites from areas of clays. These two sites are identified on the map in figure 4.9, where they can be

seen to occupy positions close to the boundary between major areas of clays and sands.

From these observations it appears that the occurrence of these plants of <u>A. hybridus</u> is influenced not only by characteristics of the soil in which it is growing, but also by characteristics of soils in the neighbouring area. A failure to take this into consideration will lead to the conclusion that the occurrence of the species is independent of soil characteristics.

Although plants of <u>A. hybridus</u> may grow on clay soils when these are close to sands, the observation of their presence (as noted in these surveys) leaves many questions unanswered. For example:

- 1) Had the plants observed on clays arisen from seeds produced by plants growing in the same site in the previous season, or had they arisen from seeds that had been transported in from adjacent areas?
- 2) If the seeds had been produced by plants growing at the same site, in which year were they produced?
- 3) Did the plants that were observed mature and set seed successfully themselves.

For these reasons, it is clear that differences in the reproductive history of the species cannot be determined from a single observation in the field. At the same time, knowledge of the reproductive performance of plants at each site would allow a more meaningful interpretation of their distribution. Thus the plants of A. hybridus that were

observed on clays may never set seed at these sites. Yet plants may appear each year at the sites if seeds are brought in from adjacent sandy soils in which plants can mature.

4.2.5.2 Discussion of the relationships among the data from survey 2

## Relationships between distribution and soil differences

The maps in figures 4.12 to 4.14 report the occurrence of each species in each of the sites sampled in survey 2. With each map there is a transparent overlay that describes the distribution of the major areas of different soils (as defined in table 4.15).

The distributions of A. powellii and A. retroflexus are not obviously associated with the distribution of soils.

The pattern of distribution of A. hybridus in this survey resembles the pattern of the same species observed in survey 1 in which this species occurred within or adjacent to areas of sandy soils.

In the extreme southwestern part of the area studied, the association of A. hybridus with sandy soils is less pronounced. The differences in this association may be related to the fact that the species reaches the northern and eastern limits of its distribution in the northeastern part of the area studied. Within the area studied, it appears that the "mean annual growing-degree-days" increase in a gradient from north-east to south-west (Webber and Hoffman, 1967 Fig. 10). Such differences in the growing season may interact with soil differences to permit the

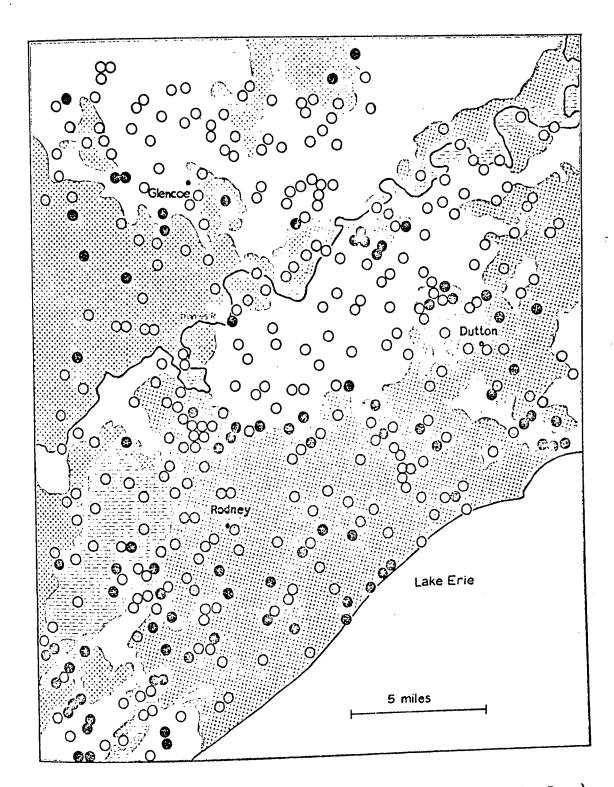


Fig. 4.12 The occurrence of A. hybridus (solid circles) superimposed upon the distribution of sites in survey 2. (See the page facing Fig. 4.9 for a key to the overlay.)

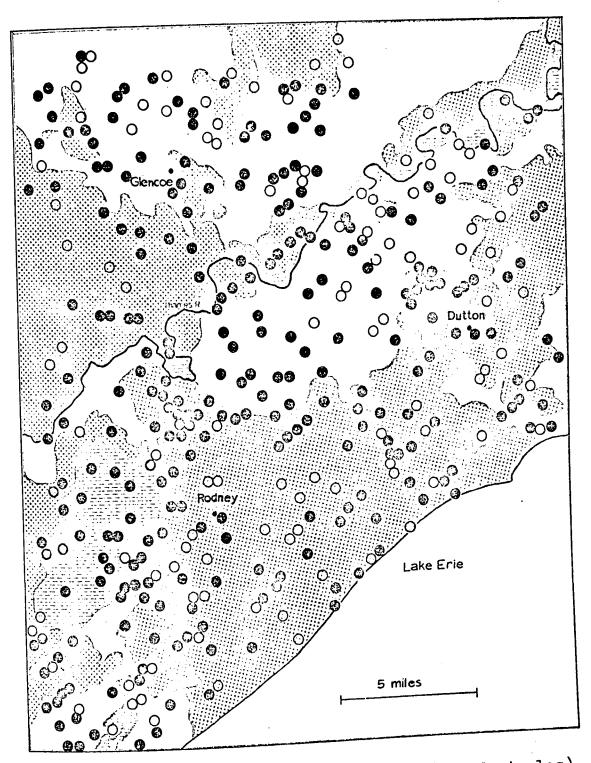


Fig. 4.13 The occurrence of A. powellii (solid circles) superimposed upon the distribution of sites in survey 2. (See the page facing Fig. 4.9 for a key to the overlay.)

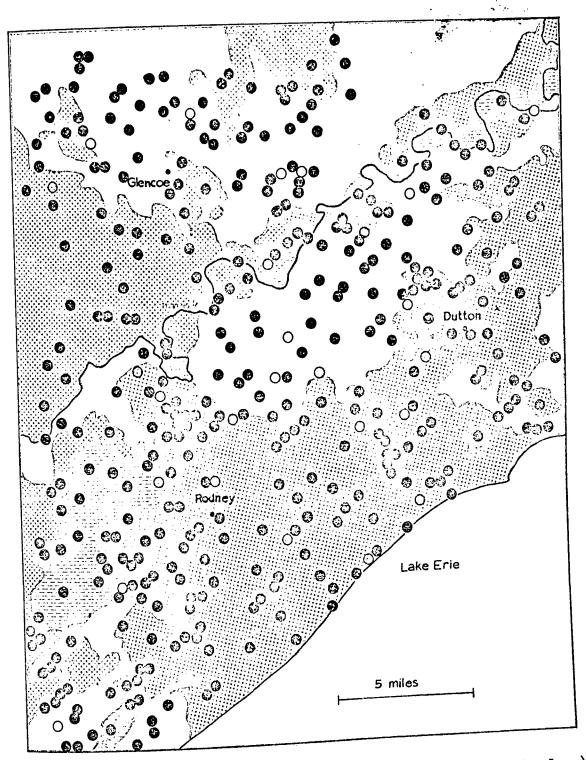


Fig. 4.14 The occurrence of A. retroflexus (solid circles) superimposed upon the distribution of sites in survey 2. (See the page facing Fig. 4.9 for a key to the overlay.)

species to set seeds successfully in a greater range of habitats in the south-west.

### 4.2.6 Conclusions

The following paragraphs (and table 4.16) contain a summary of the knowledge of the local distributions of the three species.

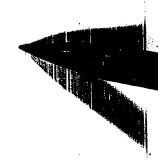
#### A. hybridus

- This was the least frequent species, occurring in 12.4% 1) of the sites in survey 1; its occurrence was not correlated with that of either of the other two species.
- This species appeared to be restricted in its distribution 2) to sites in which the soil is sandy or to sites that are close to areas of sandy soils.
- The occurrence of this species was not correlated with 3) crop type, the use of herbicides, the addition of fertilizer or the time at which the field had been ploughed.

During the course of this experimental work, records were kept of all sites in which A. hybridus was observed. of these sites were not included in the formal surveys of distribution and some came from outside the area studied. These records are of value in determining the distributional limit of the species in Southern Ontario as it appeared in the summers of 1967 and 1968. The information provided by these observations is presented in Appendix 2 in tables A2.1, A2.2 and A2.6.

### A. powellii

This species occurred in 56.3% of the sites examined in 1)

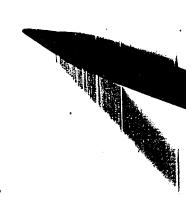


### **TABLE 4.16**

### A SUMMARY OF THE RESULTS OF STUDIES OF DISTRIBUTION

A SUMMARY OF THE RESULTS OF STUDIES	ר אָרָ	T.D.	LUT	ידד ספר.	OIN			
	Species Combinations of species							
	hybridus	powellii	retroflexus	hybridus & powellii	powellii & retroflexus	hybridus & retroflexus		
Relationship investigated	4	4	Α.	А. А.	A.A.	44		
Species differed in their frequencies				Υ*	Y	Y		
in survey 1  Species were independent in their occurrence				Y	$N_*$	Y		
Occurrence of a species was independent of the application of fertilizer								
Occurrence of a species was independent of the time of ploughing	nt Y	Y	Y					
Occurrence of a species was independe of the application of herbicides	nt Y	Y	N					
Occurrence of a species was independe of the nature of the present crop	nt Y	Y	N					
Occurrence of a species was independe of the nature of the previous crop								
Occurrence of a species was independent of the textural nature of the soil	ent :	Y Y	Y					

\*Notes: Y = Yes, the hypothetical relationship was accepted. N = No, the hypothetical relationship was rejected.



- survey 1. The occurrence of this species was positively correlated with the occurrence of A. retroflexus.
- 2) This species appeared to have no correlation between its distribution and the differences in soils throughout the area studied.
- The occurrence of this species was not correlated with crop type, the use of herbicides, the addition of fertilizer or the time at which the field had been ploughed.

### A. retroflexus

- 1) This species was the most frequent, occurring in 77.5% of the sites in survey 1; its occurrence was positively correlated with that of A. powellii.
- 2) There appeared to be no correlation between the distribution of this species and differences in soils throughout the area studied.
- The species occurred with less than the mean frequency in fields in which corn was grown or had been grown in the previous season.
- 4) The occurrence of this species was negatively correlated with the application of herbicides.
- 5) The occurrence of this species was not correlated with the addition of fertilizers or the time at which the field had been ploughed.

#### CHAPTER 5

### GERMINATION BEHAVIOUR: A REVIEW OF THE LITERATURE

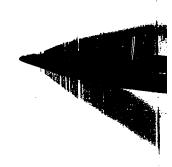
#### 5.1 Introduction

It was stated in Chapter 1 that several important aspects of the ecology of weeds involve the germination behaviour of their seeds. It may be important for the success of a weed that under some conditions its seeds remain dormant while it may be equally important that under different conditions the seeds germinate. It may be even more important that less than all of the available seeds germinate under any particular conditions.

Control of the germination of seeds is the result of interaction between inherited characteristics of the seeds and the influence of environmental variables. The nature of this interaction and of its components will be reviewed in the following pages of this chapter.

There are many examples of environmental factors that determine the course of development of plants and animals.

Often these factors are influential for a relatively short period during development, yet their effects may be evident for the lifetime of the individual. Photoperiodic induction of flowering in plants and many learnt patterns of behaviour



Therefore, as Rowe (1964) has in animals, are examples. pointed out:

"To understand why organisms are as they are, it is necessary to retrace their development, searching out along the ontogenetic route the influences that have entered into, shaped and conditioned subsequent morphology and behaviour".

The phenomenom of germination and the various factors that influence its expression can logically be examined in an ontogenetic order.

### 5.2 The Germination of seeds

Much has been written about the germination of seeds and yet seldom has the term "germination" been described. The earliest use of the term probably described events visible to the naked eye, i.e. rupture of the seed coat or coats and the emergence of the seedling. With the development of plant physiology, much has been learnt of the metabolic processes that govern the emergence of the seedling. This knowledge has lead some authors to suggest that it is the activation-processes that can be considered as For example, Mayer and Poljakoff-Mayber (1963) germination. state:

"Cell division and elongation only occur once the cells of the seed have been activated in some way, so as to permit the control of these processes by various factors. Although it appears that for visible germination only one of these processes is necessary, there can be no doubt that for normal development and growth of the seedling both cell division and It is thus possible to differentiate between these growth processes and the elongation are essential. process of activation which precedes growth and which However we will not, during the discussion of may be termed germination.

germination, attempt to make such a precise differentiation but rather will try to describe all those processes which take place upto seedling formation."

Koller et al (1962) delimit germination in the following way:

"The final process in germination is embryo growth with sufficient force to rupture whatever embryo-covers are included in the dispersal unit. The starting process is commonly considered to be imbibition, but the germination behaviour of a seed may be conditioned as far back as fertilization."

This definition apparently includes those seeds that imbibe immediately after dispersal yet do not complete their germination in the absence of other requirements which may not be met for weeks, months or years. Such seeds could be said to be germinating at all times between dispersal and seedling emergence, no matter how long the interval. This concept may be justified since the changes within a seed are probably of a continuous and quantitative nature as are many other biological phenomena. Moreover, environmental factors are acting continuously to influence both the rate and direction of after-ripening and activation-processes.

Nevertheless, in order to study the influence of environmental variables on the entire process of germination, it may be convenient to isolate different stages in this process. Three arbitrary categories can be defined. In their ontogenetic order they are: preconditioning, dormancy and the emergence of the seedling. In the following discussion, dormancy will also include after-ripening.

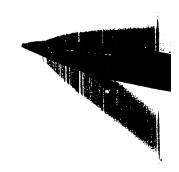
The emergence of the seedling is an important stage in

the ecological study of germination. At this stage, the embryo makes the transition from the internal environment of the seed into the external environment. Thus it loses the influence of seed and fruit coats that have buffered the extremes of variables in the external environment, particularly moisture, temperature and wilnerability to predators and parasites. If the seedling is able to synthesise new plant material, it can begin to develop resistance to the external environment in the form of new protective tissues.

Apart from its ecological importance, in experiments with large numbers of seeds emergence provides the only rapid and thus realistic indication of germination in an individual seed. In the experiments described in this thesis, environmental factors acting at earlier stages of germination have been examined for their effect on emergence. For these reasons use of the term "germination" has been restricted to the emergence of the embryo into the external environment.

### 5.3 Preconditioning

Environmental factors that influence the development of the seed before it has reached maturity on the parent-plant are described collectively as preconditioning. Since these events are the farthest removed in time from emergence of the seedling, it is not surprising that their effect on emergence has received little attention. Rowe (1964) and Evenari (1965) have reviewed the few reports that do exist.



The effects of different temperatures during maturation have been reported the most frequently. Koller (1962), working with lettuce, variety 'Grand Rapids', found a difference in germination response between seeds from plants subjected to different temperatures during the period after the onset of flowering. Plants subjected to higher temperatures (26°C constant, or alternating 26° and 17°C) produced seeds that gave a greater percentage of germination at high temperatures (23° and 26°C) than seeds produced by plants subjected to a low temperature (17° constant). Maun and Cavers (1970) have found a similar effect with Plants of this species grown in clones of Rumex crispus. soils at high temperatures (26° and 35°C) produced seeds that germinated more rapidly than seeds from plants grown in soils at low temperatures (10° and 15°C). Lipp and Ballard (1963), using clones of Anagallis arvensis, observed an inverse relationship between the proportion of seeds that were dormant and the temperature at which the parent-plant had been maintained. Sexsmith (1969) demonstrated with Avena fatua that a lower percentage of dormant seeds are produced following high temperatures during plant growth.

The effects of temperature-preconditioning on seed size in <u>Amaranthus retroflexus</u> have been recorded by McWilliams, Landers and Mahlstede (1968) but no mention was made of subsequent germination.

Koller (1962) showed that photoperiod affects

preconditioning. Plants grown under continuous illumination

produced seeds which gave a greater percentage of germination than seeds from plants grown under a photoperiod of 8 hr light and 16 hr darkness. Referring to the possibility that germinability may be negatively correlated with humidity he suggests:

"if water-stress during maturation improves germinability, the beneficial effects of continuous light may also involve increase in water stress, by prolonging stomatal opening."

Evenari (1965) examined photoperiodic preconditioning in <u>Diplotaxis harra</u>. Seeds from plants subjected to the shorter of two daylengths gave the greater proportion of germination.

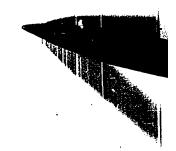
McWilliams (1966) examined the germination of seeds from clones of <u>Amaranthus retroflexus</u> grown under different moisture regimes. There were no differences in germination at 35°C; all samples of seeds gave between 95 and 100% germination. This was the only temperature at which germination was examined although seeds of this species can germinate at temperatures as low as 25°C (Barton, 1945).

Sexsmith (1969) grew plants of Avena fatua under two different regimes of moisture. Seeds maturing on plants under dry conditions were less dormant than those maturing on plants under moist conditions. The magnitude of the effect varied over a range of different temperatures.

The preconditioning effects of factors associated with the agricultural environment also have been examined experimentally. Rojas-Garciduenas and Kommedahl (1960) considered the

effects of the herbicide 2,4-D applied to plants of Amaranthus retroflexus at the onset of flowering. spite of this treatment, the plants eventually produced seeds and the germination response of these was examined at 15 days and at five months after their harvest. each time seeds from plants that had been sprayed gave a percentage of germination five times as great as seeds from unsprayed plants. Maun and Cavers (1969) examined the effect of 2,4-D applied to clones of Rumex crispus at different times during the development of flowers and fruits. Application 12 days before anthesis and at anthesis produced At 7 days after anthesis seeds with small inviable seeds. embryos and low viability were produced. Application of 2,4-D 34 days after anthesis had no effect on development of the embryos or germination of the seeds. The result of spraying in these two investigations was to modify the development of seed although the expression of this modification was a greater percentage of germination (perhaps less dormancy) in one case and lower viability in the other.

Seed production of barley has been studied following the addition of nitrogen and phosphorus fertilizers (Chiang and Robertson, 1968). At four out of five locations, the proportion of large seeds produced was greater in treatments that had received an application of nitrogen. While there was no obvious correlation between seed size and time to emergence, seedlings from larger seeds grew more vigorously



after emergence from the soil than those from smaller seeds. The addition of phosphorous was followed by no marked change in seed size or time to emergence although the nitrogen content of the seeds differed from that of seeds of untreated plants.

The internal environment of the parent-plant can undergo fluctuations that may influence maturing seeds. For example, in plants with an indeterminate pattern of growth, physiological changes may take place between the onset of flowering and its culmination. The number of seeds in different stages of maturation would vary with time and the demands these seeds would make on parental resources Even in a plant with a determinate would vary accordingly. pattern of growth, there is a strong possibility of competition for nutrients between individual ovules. Differences in the position of seeds on the inflorescence may determine the supply of nutrients to a particular site. Salisbury (1942 p.33) reviewed evidence suggesting that variability in seed-size is largely influenced by competition between flowers on the same plant and even between seeds in the same fruit.

Quinlan and Preston (1968) thinned flowers and fruitlets on apple trees to observe the effect of reducing the competition between fruits on the same plant. They found increases in the size of fruit and the cross-sectional area of the trunk that could be attributed to an increase in meristematic activity. They suggested that competition was

for a substrate important in the control of meristematic activity. Their results support the view that competition is most intense at the time of blossoming and gradually diminishes during the following three to four weeks.

Cavers and Harper (1966) examined the germination response of seeds of Rumex crispus and R. obtusifolius produced at different positions on the parent plant. Germination was examined under three light and temperature regimes; continuous darkness and constant temperature, continuous darkness and alternating temperatures, and alternating light and darkness (with alternating temperatures). Seeds were compared from (a) different panicles on the same plant, (b) upper and lower positions on the same panicle, and (c) positions distal and proximal to the main axis on the same branch of the panicle. In alternating light and darkness, all of the seeds germinated from any position. In darkness, with either constant or alternating temperatures, seeds from different panicles gave different percentages of For most panicles, fewer seeds from upper germination. parts of the panicle were dormant than from lower parts. Seeds from the base of the branches of the panicle proximal to the main axis were less frequently dormant than seeds from the distal to the main axis ends.

Both Rowe (1964) and Evenari (1965) have raised the question of the earliest stage in the life of the parent plant at which environmental factors can influence the eventual germination of seeds. Most of the experimental

work outlined so far has employed treatments either during the entire lifespan of the parent-plant or merely during the period after anthesis. Evidence is accumulating of environmental factors that have a carry-over effect on the phenotype of first generation offspring and even on offspring of subsequent generations. Although these effects have been noted on such characters as plant height (Highkin, 1961) and yield (Went, 1959 and Durrant, 1958), their implication for germination response is important. Highkin has suggested that the influence of previous environmental conditions may be inversely proportional to the number of generations that separate the particular environment from the generation in which the effect is observed.

The extent of preconditioning described above may have important implications for experimental work. Thus plants grown under uniform conditions in order to observe genetic differences or the effects of different treatments may exhibit variability that is a function of environmental influences in preceding generations. Rowe (1964) has discussed the implications of these phenomena for forestry research and particularly for the technique of provenance—testing.

### 5.4 Dormancy, after-ripening and emergence

Several authors have drawn attention to the confusion that exists in defining dormancy. (Thurston 1960, Evenari 1965, McWilliams 1965). Thurston (1960) considered Harper's

Harper recognised three categories of dormancy; innate, induced and enforced. Innate dormancy is a genetically controlled property of the ripe seed as it leaves the plant, and it is only "broken" when the seed has experienced certain internal or environmental conditions. Induced dormancy develops in a non-dormant seed in response to certain environmental conditions and continues when these conditions no longer prevail. Enforced dormancy describes the situation when seeds are prevented from germinating simply because the particular conditions in the external environment will never permit germination. When unfavourable conditions cease to prevail the seeds germinate.

Evenari (1965) defined the term dormancy as partly blocked and controlled germinability. He went on:

"When these blocks are inactivated either by their physiological removal or by their circumvention, dormancy is broken' i.e. the dispersal units passed [sic] from the 'dormant' to the 'non-dormant' state as happens e.g. during after-ripening."

Evenari (1965) went on to justify use of the term "controlled germinability":

"When dormancy is looked at from the aspect of its biological importance it is obvious that if fulfils

1) It enables the dispersal unit to survive adverse external conditions. This is possible because during dormancy the resistance of the dispersal unit to the damaging influence of extreme external conditions is much heightened.

2) It restricts germination in time and place and permits it only in certain habitats, soils, light conditions, certain seasons of the year etc. where and conditions, certain seasons of the germinated when the probability of survival of the germinated when the probability of survival of the germinated when the probability of survival of the germinated experimentally seedling is greatest. This has been proved experimentally in many cases by myself and Koller and his collaborators. This is why we may talk of 'controlled germinability'."

Evenari (1965) considered dormancy to be of two basic types; dormancy originating within the embryo and dormancy imposed upon the embryo by some agent external to it. The source of imposed dormancy may be the fruit coat, the seed coats, the endosperm or the 'pulp'. Such dormancy may be the result of any of the following conditions, acting independently or in combination:

- 1) Impermeability to water.
- 2) Impermeability or differential permeability to certain gases.
- 3) Mechanical opposition to the expanding embryo.
- 4) Presence of germination inhibitors.
- Where dormancy is imposed upon the embryo by other tissues in the dispersal-unit, removal of these, and particularly the fruit or seed coat, allows the embryo to germinate.

  When the isolated embryo fails to germinate due to physiological blocks within the embryo itself, this is described as embryo dormancy. It may arise for the following reasons:
  - 1) Presence of substances inhibitory to germination.
  - 2) The embryo at dispersal time is structurally immature.
  - 3) The embryo at dispersal time is physiologically immature. Evenari (1965) rejected Barton's (1945) "dormancy in the

Evenari (1965) rejected Barton's (1947) documents dry state" as a form of dormancy. He considered that seeds that only require water to germinate are non-dormant. He would probably consider Harper's (1957) 'enforced dormancy' a case of non-dormancy.

Most discussions of dormancy appear to have been directed towards the goal of obtaining an absolute term (e.g. dormant or non-dormant) that can be used indisciminately to describe a particular collection of seeds. Little consideration has been given to the varied and varying conditions that individual seeds naturally encounter between the times of dispersal and germination, and to the different degrees of dormancy exhibited in samples of seeds under such conditions. An exception is Vegis (1963, 1964), who places dormancy and germination within their ecological context as adaptations for survival. He states:

"Through protracted selection under natural conditions, different plant forms have become precisely adapted to various climatic conditions. Under the influence of certain conditions which regularly precede the unfavourable season of the year, metabolic processes in the shoot apex and in the embryo switch from growth to decreasing activity and then dormancy, with the simultaneous development of high resistance. The decrease of growth activity both in the embryos and in the buds of perennial plants manifests itself in lowered capacity to react immediately by continued growth to certain external conditions which are growth-promoting during the active phase."

There is an indeterminate number of individual variables that can contribute to the external environment of a plant. These variables interact to produce an unknown but even greater number of different permutations of environmental conditions. In response to this variety, it is little wonder that many different expressions of seed dormancy have evolved. Vegis (1963, 1964) described in detail the relationships between changes in growth activity in the seed and the temperature range within which germination can

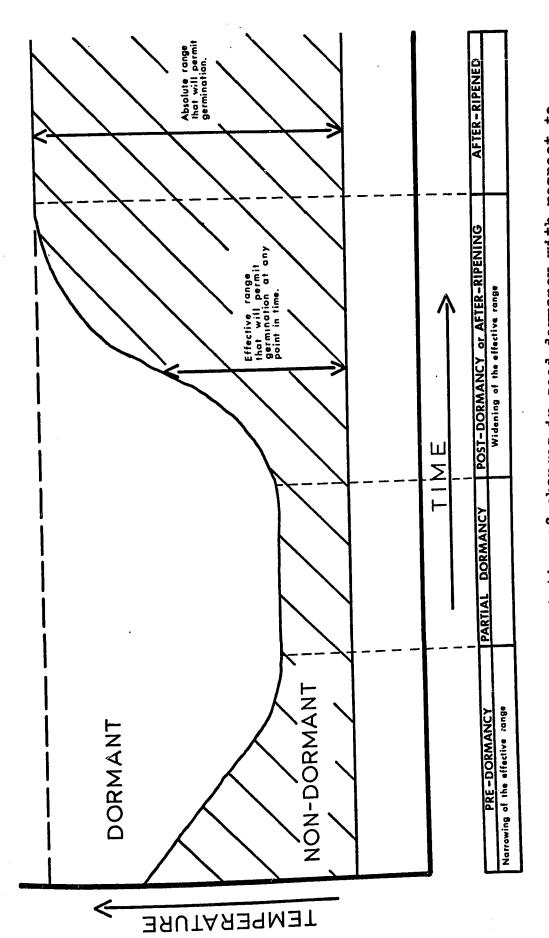
occur. Although the specific effects of temperature on seed dormancy and germination will be discussed later, Vegis (ibid) developed a useful way of describing dormancy and this will be presented now.

With respect of temperature, there is an absolute range within which germination can occur depending upon the state of other environmental variables (Fig. 5.1).

For a specific set of conditions, there is, within the absolute temperature range, an effective range of temperatures within which germination does occur. Changes that occur during dormancy do not influence the absolute temperature range, but they may influence the effective temperature range. Such changes reflect the state of "growth activity" or metabolic activity in the embryo.

As a seed matures upon the parent-plant, growth activity usually declines. While growth is active the effective temperature range that promotes germination is wide. As growth activity declines the effective range narrows; this is termed "pre-dormancy".

In a sample of seeds, narrowing is first indicated at a temperature close to the limit of the effective range. At this temperature, less than 100% of the viable seeds germinate. As growth activity continues to decline the proportion of seeds that germinate at the same temperature falls until the time is reached when no germination occurs. This temperature can then be said to be outside the effective range that will permit germination. In this way the



A graphical representation of changes in seed dormancy with respect to temperature and the age of the seed (Based on Vegis, 1963, 1964). F18. 5.1

effective range of temperatures gradually narrows.

The ecological significance of this behaviour is that germination is prevented before or during a period of adverse conditions for seedling establishment or plant growth. The effective temperature range that will permit germination continues to narrow until it excludes those temperatues that characteristically precede the unfavourable period or prevail for its duration.

Theoretically this is a sufficient precaution and there is no need for germination to be inhibited at temperatues that are unlikely to occur. Nevertheless, this situation is described by Vegis (1963, 1964) as "partial dormancy".

In some species narrowing of the effective temperature range continues until all temperatues are excluded.

Germination is then completely inhibited and the situation is described as "true dormancy".

The restricted range of temperatures that permit germination may be maintained for many months, but eventually a reversal of the trend is initiated; activity increases in the embryo and is accompanied by a widening of the range of temperatures at which germination can occur. The range—widening process is termed "post dormancy" and is equivalent to after-ripening.

There is no reason why this concept should not be extended to include other environmental variables if it can be shown that they elicit a similar response. Although none have been examined in precisely the same way, there is

evidence that suggests similarities. For example,

Evenari (1965) listed three of the main characteristics

of after-ripening as:

- "1) The widening of the temperature range over which the seeds are able to germinate.
- 2) The loss of photo-requirement and the increase in photo-sensitivity.
- 3) The gradual increase in germination velocity."

#### He added a fourth:

"The sensitivity towards germination inhibitors decreases during after-ripening."

Evenari (ibid) went on to state that:

"After-ripening is qualitatively and quantitatively influenced by external conditions such as temperature, air humidity, light conditions etc."

If the germinability of seeds is plotted graphically for one environmental variable against time, as in figure 5.1, a two-dimensional dormancy profile is obtained with If the respect to that particular vaiable and time. interaction of the first variable with a second variable is considered, a three dimensional profile may be obtained. This procedure could be extended, at least theoretically into multi-dimensional space to include all the internal and external variables known to influence the entire germination The result would provide an accurate description process. of dormancy in the quantitative terms of each variable If this technique were adopted, any statement considered. about the dormancy of a sample of seeds could be qualified by the conditions under which the assessment was made.

With respect to any one specific set of environmental

conditions, the response of a seed may be described under the following categories:

- 1) The seed germinates in the shortest known time for these conditions; it is non-dormant.
- 2) The seed ultimately germinates under the conditions in question, but there are other requirements that must be met before it will do so; it is dormant.
- The seed will never germinate under these conditions.

  It is inappropriate to describe this situation in terms of dormancy, although many of the phenomena described as enforced dormancy fall within this category. At the same time, these conditions may influence substantially the subsequent germination responses of viable seeds under other sets of conditions.
- 4) The seed is killed under these conditions.

The response of an individual seed to certain conditions will be as in 3) or 4) above, depending upon inherent characteristics of the seed or upon preconditioning. This is a reason for including inherent variables and variables that contribute to preconditioning in the matrix of conditions that describe dormancy.

If dormancy is described with respect to the entire range of environmental variables, or even to a defined number of these, it becomes evident that under some sets of conditions a seed may be dormant while at the same time it would be non-dormant under different sets of conditions.

In describing a germination response, the most frequent

parameter measured is the percentage or proportion of emergent embryos in a sample of seeds of known size.

Differences in the percentage germination do not describe the degree of dormancy of an average seed in the sample, but rather the frequency of dormant and non-dormant seeds in the sample. The figure reported is a measure of the probability that a seed chosen at random will be non-dormant. It is naturally impossible to describe the germination profile of an individual seed once it has given a positive response. Thus it is necessary to use large samples and work in terms of probabilities.

Another parameter encountered is the rate of germination. This is really a frequency distribution of values for the parameter "time to germination" among those seeds that are non-dormant or possess a dormancy that is lost after a period of time in the conditions in question.

# 5.5 Environmental factors controlling germination 5.5.1 Preamble

There have been many extensive reviews of the inherent and environmental factors controlling germination (Crocker, 1948, Crocker and Barton, 1953, Harper, 1957, Mayer and Poljakoff-Mayber, 1963, Vegis, 1963 and 1964, Evenari, 1965, McWilliams, 1965 and 1966). Most of these authors consider environmental factors individually or as interactions between a small number of factors, and realistically this is the most one can expect. Nevertheless a recurring weakness in

much experimental work is a failure by authors to define precisely the state of those environmental variables that are held constant (or at least assumed constant). Since independent investigations of identical factors in the same species are doubtless conducted under different experimental conditions, a failure to report these conditions results in a loss of information that would be available from comparison between investigations. These problems have been considered in detail by Lawrence et al (1947).

In preparing a further review of factors controlling germination the procedure will be to briefly outline the range of effects of a particular factor and to enter into specific details only when the results relate to Amaranthus or to the specific kind of experiment described subsequently in this thesis.

5.5.2 Temperature as a factor controlling germination

One factor that fluctuates most frequently in the plant environment is temperature. There are both high temperatures and low temperatures that place limits on the performance or survival of plants. In temperate regions there are protracted periods of low temperatues that are unfavourable for the survival of some species and it is the function of the seeds to maintain the species throughout these periods.

Periods of time that are unfavourable for other reasons (e.g. the availability of moisture) may also be associated with characteristic temperatures. Daily temperatures

fluctuate between upper and lower levels which also follow a predictable cycle of fluctuation over the period of a year. Periods of unfavourable conditions frequently occupy a characteristic position in the annual cycle of temperature fluctuations. Thus the temperatures that prevail before and after the unfavourable period also are predictable. This situation provides the selection pressure for the evolution of responses to temperature that result in the control of the timing of germination.

Seeds of different species have different temperature ranges within which they germinate (Mayer and Poljakoff-Mayber 1963). Even within species the temperature range for germination varies between ecotypes. McWilliams, Mahlstede and Landers (1968) working with Amaranthus retroflexus collected from most of its geographical range in North America, found that seeds from plants grown under identical conditions at Ames, Iowa had a germination response at 20°C that was correlated with the latitude of the original collection.

Within a temperature range the optimum has been defined as that temperature at which the highest percentage of germination occurs in the shortest time (Mayer and Poljakoff-Mayber, 1963). According to Vegis (1963), a germination profile with a distinct peak percentage of germination at an optimum temperature and a declining percentage germination with higher and lower temperatures is a sign that after-ripening is incomplete. When after-ripening

is complete all or nearly all the seeds germinate at any temperature within the final range. Non-afterripened seeds may give maximum germination at high or low temperatures depending upon the species (Vegis 1963).

Evans (1922) recorded the rate of germination of drystored seed of Amaranthus retroflexus at temperatures between She found that germination proceeded to near 9° and 42°C. Below this completion in any temperature above about 15°C. temperature, low levels of germination were obtained and then only if the seeds were scarified. Throughout the range of temperatures studied the speed of germination increased with higher temperatures. Kadman-Zahavi (1960), using drystored seeds of the same species, found a similar relationship between temperature and germination rate. germination percentage that he recorded was also proportional This pattern of response indicates that the to temperature. seeds that he used may not have been fully after-ripened.

### Alternating temperatures

In most experimental investigations the effects of constant temperature treatments have been examined. The validity of interpretations based on these results may be questioned since natural temperatues usually fluctuate within a daily cycle. Vegis (1964) raises this objection and then adds:

"However, it is established that in order to exert its full maximum effect on the growth activity, a semperature need not act continuously. Several hours a day may be satisfactory, provided that the temperature of the other part of the day does not act in a contrary direction."

With specific respect to the effect of alternating temperatures on seed germination Vegis concludes:

"in this case, one of the two temperatures is usually the most [sic] critical and must persist for the greater part of the day."

The temperature of the soil may show diurnal fluctuations to a depth of one metre (Daubenmire 1967) and thus seeds that are buried may experience day to day temperatures that are almost constant depending on the depth at which they are buried. There is no evidence that seeds of species of Amaranthus will germinate below two inches and thus it is unlikely that seeds of these species will naturally germinate in response to constant temperatues. However, constant temperatues may have an important role in preventing the germination of seeds buried at great depths.

McWilliams (1965) compared the germination of seeds of several species under different constant and alternating temperature regimes. In incubators maintained at different constant temperatues, samples of seeds of Amaranthus retroflexus gave germination percentages that were directly related to temperature. The percentages ranged from 20 at 68°F to 79 at 95°F.

In order to compare germination under alternating temperatures, each temperature regime was described in terms of heat units per day. These were obtained by summing the products of temperature and the number of hours at that temperature for both temperatures in the cycle. The results for seeds of A. retroflexus indicated a direct relationship between heat units per day and percentage germination in all

but one treatment. The exceptional treatment was an alternation between 10° and 35°C, a greater range than in any other treatment. Thirty-nine percent germination was observed in this treatment which was calculated as 6500°-hr per day. The percentage of germination in other treatments ranged from 31 for 6700°-hr to 53 for 9500°-hr.

After attempting to correlate germination under constant and alternating temperature regimes, and also to correlate germination in incubators, the greenhouse and the field,

McWilliams (1965) stated:

"There are several diverse temperature regimes which will result in either very high or very low germination percentages of Amaranthus and Portulaca. The explanation of the germination of these species under field conditions depends upon an understanding of the effects of alternating temperature on germination."

## Fluctuations in the range of temperatures that promote germination

Vegis's (1963, 1964) concept of dormarcy in terms of the temperature ranges that permit germination, has been discussed on pages 167 to 170. The exact form that the narrowing of each range takes varies from species to species. Vegis (1964) recognised three different patterns of change. Each of these will now be discussed.

# a) A narrowing of the range to restrict germination to low temperatures

As metabolic activity declines, germination is restricted to progressively lower maximum temperatures. When narrowing of the range ceases, germination may be restricted to a narrow range of lower temperatures or where this process has continued further, germination may not occur at any

temperature. After a period in which the range remains constant, the initial trend is reversed and germination becomes possible at progressively higher temperatures.

This pattern of temperature response is characteristic of plants that produce seeds before the onset of a hot dry season inhospitable to seedling growth. The foremost need of such seeds is to avoid germination in response to the temperatures that prevail before and during the hot season. Examples of species who seeds show this response are:

Vaccaria pyrimidata and Agrostemma githago (Boriss, 1940, Boriss and Arndt, 1956) and Veronica hederifolia (Wehsarg, 1918).

## b) Narrowing of the range to restrict germination to high temperatures

With seeds that exhibit this pattern of response, germination is restricted to progressively higher minimum temperatures. A subsequent widening of the range occurs as seeds are able to germinate at progressively lower minimum temperatures. Species that have seeds with this response appear to be adapted to a climate with a regularly recurring cold season. The production of seeds in such species is normally completed in the autumn when further high temperatures are unlikely to prevail. Examples of species in this category are: Amaranthus retroflexus (Crocker, 1916), Thlaspi arvensis (Wehsarg, 1918) and Betula spp. (Joseph, 1929, Meiss, 1926).

Crocker (1916) observed that ripe seeds of Amaranthus retroflexus, freshly harvested from green plants, could

only be induced to germinate at temperatures above 40°C. As after-ripening progressed the seeds germinated at progressively lower minimum temperatures. Barton (1945), working with the same species, compared the germination at different temperatures of freshly harvested seeds and seeds held moist under conditions of darkness at 20°C for two years. The results of these germination tests are presented in table 5.1.

TABLE 5.1

EFFECTS OF TEMPERATURE ON THE GERMINATION OF FRESHLY HARVESTED AND MOIST STORED SEEDS OF AMARANTHUS RETROFLEXUS, 1942 CROP

(From Barton (1945) table 3)

Percentage germination after moist storage at 20°C for:

Germination temperature °C	O de Collection	at 20°0	(a ) A	days Collection B
Constant  15 20 25 30 35	0 2 18 16 86	0 0 8 11 73	0 0 84 86 91	1 0 91 90 90
Alternating 10/20 10/30 15/30 20/30	0 0 1 1	0 2 6 3	21 83 84 85	12 90 83 88

These results can be interpreted according to Vegis's (1963, 1964) concepts of dormancy. When freshly harvested, the seeds have already completed their pre-dormancy and

germination is restricted to high temperatures in most seeds. For germination at constant temperatures, after-ripening appears to be complete after two years of moist conditions. Similar interpretations can be placed on the results observed under alternating temperatures. The small percentage of germination observed at 10°/20°C indicates that either after-ripening is incomplete, or polymorphism exists among the seeds with respect to their ability to germinate under these conditions.

Barton (1945) also examined the germination of seeds from the same sources after periods of dry storage. A gradual change took place in the capacity of these seeds to germinate at different temperatures. After two or three months they were able to germinate over a wide range of temperatures, including 20°C. Seeds that had been stored in moist conditions for two years did not germinate at 20°C. The comparison between moist-stored and dry-stored seeds suggests that under moist storage conditions, after-ripening is not complete even after two years. These results are discussed further on page 183.

## c) Narrowing of the range to restrict germination to intermediate temperatures

Vegis (1964) considered this to be the most common pattern of response of seeds to temperatures. As metabolic activity declines, germination is restricted to both progressively lower maximum temperatures and progressively higher minimum temperatures. The ability to germinate is maintained, if at all, at intermediate temperatures.

Eventually a widening of the temperature range takes place as the seeds are able to germinate at higher maximum temperatures and lower minimum temperatures. This pattern of response appears to be an adaptation to climates with two unfavourable seasons; a hot arid summer and a cold winter. Examples of species whose seeds exhibit this response are: Rumex crispus (Wehsarg, 1918) and Chenopodium album (Krug, 1929).

### d) Temperature as a factor influencing the temperature range for germination

The rate at which a seed develops dormancy and then loses this dormancy through after-ripening is controlled by various external factors including ambient temperature. Generally, high temperatures cause a narrowing of the temperature range for germination while low temperatures favour a widening of this range (Vegis, 1963). This effect of low temperatures is know as stratification and it has been demonstrated by Taylorson and Hendricks (1969) with Amaranthus retroflexus. Seeds of this species that had imbibed in darkness at various temperatures were put to germinate at 35°C in darkness. Less than 5% germination was observed for seeds that had imbibed at temperatures above Greater percentages of germination followed 20°C. imbibition at temperatures from 5° to 20°C, with an optimum The number of days in imbibition at a value at 15°C. particular temperature required for the full expression of the response increased with temperatures both lower and higher than 15°C.

#### Secondary dormancy

High temperatures during post-dormancy sometimes induce a second narrowing of the temperature range that will permit germination of those seeds that are not fully after-ripened (Vegis 1964). Courtney (1968) has shown that seeds of Polygonum aviculare that have not germinated by May become dormant once again. This secondary dormancy is only broken after another winter of low temperatures. He was able to induce secondary dormancy experimentally by incubating at 25°C seeds that almost had completed their after-ripening at 4°C.

Koller (in discussion following Vegis, 1963) questioned whether primary and secondary dormancy are identical since several instances are know in which seeds in secondary dormancy and seeds in primary dormancy require different treatments before germination can take place.

## Periodic fluctuations in the temperature range that will permit germination

The experimental work of Barton (1945) with Amaranthus retroflexus has been described in part on page 181 and is also described on page 197. The discussion of the germination patterns of the seeds stored in moist conditions at 20°C for two years is not complete unless several other observations are taken into account. During the two years of storage at 20°C a certain number of seeds did germinate within well defined periods. The first period of germination began after 10 months and lasted for about 3 months whereupon there was little germination until the 22nd

month of storage. This exhibition of periodicity in germination raises an interesting question. Had the seeds that germinated been prevented from so doing, perhaps by storage at lower temperatures, would they have retained the ability to germinate at 20°C after the 13th month? And if they had lost the ability would this situation be defined as secondary dormancy or merely a continuation of the after-ripening process? Considering the seed sample as a whole, after-ripening can be said to have been incomplete since a measure of control over germination at a particular time and temperature was maintained.

### The interaction of temperature with other factors

Interactions between the effects of temperature and light on seed germination are included in the reviews by Vegis (1963, 1964). His own work with Betula verrucosa provides an example. Partly after-ripened seeds germinate in darkness only within the temperature range 20° to 30°C. When germination tests are performed in the light, seeds will germinate at temperatures between 10° and 30°C.

There are also species which exhibit the opposite response, i.e. a narrower range of temperatures that promote germination in the light than in darkness. Vegis (1964) includes Amaranthus spp. in this category.

Interactions between temperature and the spectral quality of light have also been reported. Taylorson and Hendricks (1969) determined the duration of dark imbibition required to allow maximum germination of seeds of Amaranthus

retroflexus following a red light treatment. The seeds imbibed at temperatures between 5° and 35°C and germination tests were conducted at 35°C. They found that the duration of imbibtion required for maximum germination varied inversely with temperatures; at 35°C, 36 hr was sufficient, whereas at 5°C more than 5 days was necessary. Illumination with red-light stimulated more germination than in controls that remained in darkness. However, the effect of red-light on the germination response could be nullified by exposure to far-red light.

Four different factors, in all of their combinations, were examined for their effect on germination of the seeds of three different species, in the greenhouse (Wiese and Davis, 1967). One of the species was Amaranthus retroflexus. The factors included were; four different alternating temperature regimes, two moisture treatments, two soils and five depths of sowing. The temperature regimes, each for a 16-hour day and 8-hour night were: 10° and 2°, 18° and 10°, 27° and 18°, and, 35° and 27° (all °C). Sowing depths were 0, ¼, ½, 1, 2 and 4 inches below the soil surface.

For seeds buried at ½ and 1 inches, highest germination was at 27°/18°C. Seeds buried at ¼ inch gave equal amounts of germination over the temperature range 18°/10° to 35°/27°C and seeds buried at 2 inches and below gave less than 10% germination at any temperature. Seeds on the soil surface germinated less than seeds buried at ¼, ½ and 1 inches at

### TABLE 5.2

#### A SYNOPSIS OF PREVIOUSLY REPORTED DATA DESCRIBING THE TEMPERATURE REQUIREMENTS FOR GERMINATION OF SEEDS OF AMARANTHUS

Author	Species	Storage of a germinati Period of storage
Crocker (1916)	A. retroflexus	nil
Evans (1922)	A. retroflexus	6 to 18 months
Barton (1945)	A. retroflexus	nil 24 months 2 to 3 months
Rojas-Garciduenas & Kommedahl (1960)	A. retroflexus	3 to 8 months
Kadman-Zahavi (1960)	A. retroflexus	not stated
McWilliams (1965)	A. retroflexus	5 to 6 months
Weise & Davis (1967)	A. retroflems	not stated
Notes: 1 - 16.3°C 2 - 11.9°C 3 - about 9	a) 20 6 <b>-</b> alter	nating temperature /10 b) 30/10 c) 30 nating temperature /15 b) 30/15 c) 30
4 - 14.5°C	7 - alter a) 10	nating temperature /2 b) 18/10 c) 27

TA NTS HUS

rminatio	eds before n trials Conditions	Experimental treatments	Perc 10	foll	.owi	ng te	ermin emper 30	ratu:	on res 4
	~~~~~~~~~								G.
nths	not stated	a) nil b) ground with sand c) treated with H <sub>2</sub> SO <sub>4</sub>	30 <sup>4</sup> 16 <sub>4</sub> 80	501 962 77	84 99 86	82 100 86	94 98 94	96	9i 9i 10i
ths	moist 20°C dry	nil nil nil		0	2 0 GR	18 84	16 86	86 91	
ths	not stated	a) nil b) 50 ppm 2,4-D for 60 sec				42 70			
	dark bottles	a) darkness b) 60 sec illuminatio	n		30	31 92	36	44 94	
ths	not stated	a) light b) darkness nil	2	0	20	46 34	47	79	
·	not stated	a) on soil surface b) buried ¼ inch c) buried ½ inch d) buried 1 inch e) buried 2 inches f) buried 4 inches							
ratures c) 30/	of 15 d) 30/20	8 - GR = germination percentage n	record ot sta	ed b ted	ut				
ratures c) 30/2	of 20 d) 35/10								
ratures c) 27/	of 18 d) 35/27								

ıl	Perc	foll	owir	ig te	emner	natio atur 35	200	a	ъ	c	ď
<del></del>		·					GR <sup>8</sup>		100	tnoi	tes)
sand h	304 162 80	501 962 77	84 99 86	82 100 86	94 98 94	96		7		•	
		0	2 0 GR	18 84	16 86	86 91		0 <sup>5</sup> 21 <sup>5</sup>	0 83	1 84	1 85
D				42 70							
nination			30	31 92	36 47	44 94					
	2	0	20	46 34	47	79			40 <sup>6</sup>	53	
face th th th thes thes								777777 4277	3 28 20 12 2 3	7 32 48 35 2	12 30 37 27 8
nation recorded but stage not stated											

# Responses to differences in light intensity

The light intensity in a seed's habitat may vary for a number of reasons; the amount of cloud cover, the changing leaf canopies of growing plants, the depth of soil or organic matter covering the seed and the possible remains of floral structures or fruit coats. Zahavi (1960) reported the germination rate of seeds of Amaranthus retroflexus exposed to continuous white incandescent light at different intensities. During the first two days of the trial, germination was greatest in darkness and inversely related to light intensity in the other treatments. At ten days germination was virtually complete for seeds exposed to light intensities in the range 8 to 150 foot-candles. Some seeds exposed to intensities of 2, 500 or 1000 foot-candles remained ungerminated but viable at this time.

# Responses to differences in light duration

The effects of light may be accomplished through extremely short exposures. Kincaid (1935) found that as little as 0.01 seconds of sunlight were sufficient to stimulate germination in seeds of tobacco. Seeds of Amaranthus retroflexus gave upto 50% germination in darkness (Kadman-Zahavi, 1960). 60 seconds of white light given at various times after sowing resulted in an increased percentage of germination as compared with the performance in darkness. Maximum stimulation resulted from illuminations given three days after sowing. Sauer and Struik (1964)

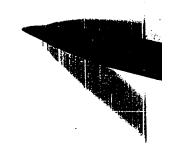
commented on the possible ecological relationship between soil disturbance, light-flash and germination. Amaranthus tuberculatus occurs within a list of species that they cite.

Mayer and Poljakoff-Mayber (1963) listed several species that have been shown to require alternating periods of light and darkness for the germination of their seeds. Frequently these requirements are modified according to the temperature at which germination is examined. In <u>Escholtzia argyi</u>, for example, the highest germination was obtained when a 6-hour photoperiod at 25°C was followed by an 18-hour thermoperiod at 5°C (Isikawa and Ishikawa, 1960).

# Responses to differences in light quality

Seed germination is one among many growth responses that are controlled or influenced by the spectral quality of light through the agency of phytochrome (Hendricks and Borthwick, 1963). Red light usually promotes germination while blue and far-red are usually inhibitory (Mayer and Poljakoff-Mayber, 1963). Resuehr (1939) reported the effects of light of different wavelengths on the germination of seeds of Amaranthus caudatus. Inhibition of germination occurred at 4500%, between 4750 and 4900%, and between 7000 and 7500%, while promotional wavelengths were 6400 and between 6750 and 6800%. The effects of red light were mullified by far-red illumination and vice versa with the nature of the last illumination determining the germination response.

Seeds of <u>Amaranthus retroflexus</u> have recieved attention in studies of the action of red and far-red light on the



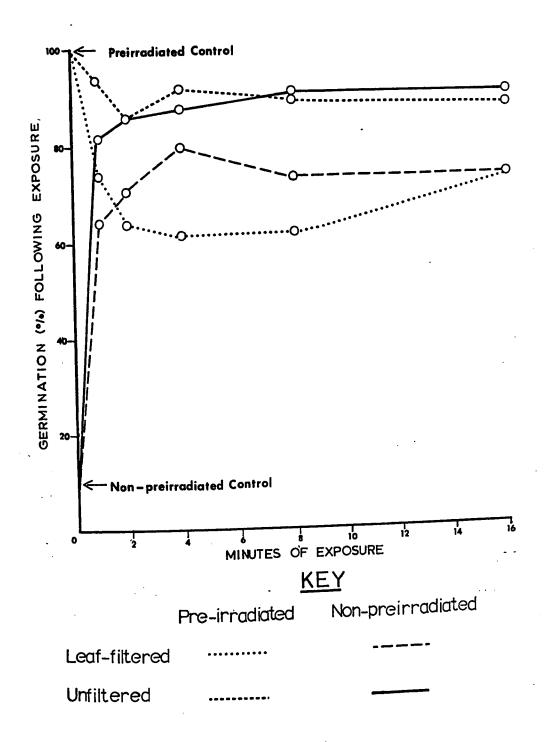
germination response. As described earlier, Kadman-Zahavi (1960) discovered that the percentage of seeds that germinated in darkness (from 30 to 44% depending on temperature) could be reduced when seeds were exposed to continuous illumination with white light of various intensities. He determined that far-red light was an In contrast, for active component in this inhibition. seeds that had imbibed in darkness a few seconds of white Complete or red light stimulated further germination. germination was observed when this stimulation was given three or more days after the beginning of imbibition. response could be modified by following the stimulatory exposure with an exposure to far-red light. When exposures to red and far-red light were alternated, each could be shown to nullify the effect of the previous exposure. Prolonged exposure to far-red was found to suppress sensitivity to subsequent red light. Sensitivity was restored only after a prolonged period of darkness.

Taylorson and Hendricks (1969) performed experiments that help to explain these responses to red and far-red light, and in particular suggest a reason for the delayed sensitivity observed by Kadman-Zahavi (1960). They concluded that the two types of phytochrome,  $P_r$  and  $P_{fr}$ , exist in the seed in dehydrated forms. The responses to red and far-red light involve the action of the phytochromes, and the time required for their hydration during imbibition is responsible for the delayed sensitivity, which they observed also.

Taylorson and Borthwick (1969) stressed an ecological implication of the effects of red and far-red light on germination. As mentioned above, germination is inhibited when the  $P_r/P_{fr}$  equilibrium shifts in favour of  $P_r$ . Seeds in different situations may receive light that differs in its relative energy levels at the red and far-red wavelengths. This will result in differential conversion rates of  $P_r$  to  $P_{fr}$  and vice versa, and thus give rise to different germination responses. Chlorophyll absorbs greater quantities of red light than far-red light. Thus light that is filtered through a foliar canopy will have a lower ratio of energy at red wavelengths to energy at far-red wavelengths. This will result in the conversion of more  $P_{fr}$  to  $P_r$  and will tend to inhibit germination.

The preceding hypothesis was tested experimentally. Seeds of several species, including Amaranthus retroflexus, Seeds of each species were divided into two were used. samples for pre-treatment. The first of these samples received 5 minutes of red light (= "preirradiated") that was intended to poise each seed in a "relatively high state of germinability". Seeds of the second sample were kept in darkness (= "non-reirradiated"). Subsequently seeds of each sample were subjected to several different treatments. The treatments consisted of exposures to various durations of either unfiltered light or light filtered through a Some seeds of each pre-treatment were tobacco leaf. The results maintained in darkness to serve as controls. of the experiment can be summarised as follows:

- 1) Tobacco leaves were found to reduce the ratio of energy at 6500Å to energy at 7300Å for sunlight, incandescent and fluorescent light that passed through them.
- 2) Seeds that were preirradiated with red light and maintained as controls gave a greater percentage of germination than seeds from non-preirradiated controls (see figure 5.2 for values for A. retroflexus).
- Preirradiated seeds that received exposures to either unfiltered or leaf-filtered light gave a smaller percentage of germination than seeds from preirradiated controls.
- 4) Non-preirradiated seeds that received exposures to either unfiltered or leaf-filtered light gave a greater percentage of germination than seeds from non-preirradiated controls.
- The final levels of germination in (3) and (4) were almost the same for seeds of the same species receiving exposures to light of the same quality (unfiltered or leaf-filtered). These ultimate percentages of germination were generally higher among seeds that received unfiltered light than among seeds that received leaf-filtered light.
- 6) In Amarenthus retroflexus seeds from preirradiated controls gave 100% germination, seeds exposed to unfiltered or leaf-filtered light gave about 90% and 70% germination respectively, regardless of pre-treatment



(Redrawn from Taylorson and Borthwick 1969)

Fig. 5.2 The total germination of seeds of Amaranthus retroflexus following exposures of different durations to leaf-filtered and unfiltered light both with and without preirradiations of red light.

- and seeds from non-preirradiated controls gave about 10% germination.
- 7) The length of exposures to the unfiltered and leaffiltered light influenced the germination response,
  particularly with exposures of 4 minutes or less.
  With exposures shorter than 4 minutes, the shorter the
  exposure the closer the germination response was to
  that of seeds of the appropriate control.

The results for seeds of Amaranthus retroflexus are summarised in figure 5.2. These results help to explain the observation that the germination of weed seeds is largely suppressed once a crop has developed a canopy of leaves that covers the soil completely.

# Changes in the response to light during after-ripening

Evenari (1965) lists as one of the features of afterripening the loss of photorequirement and an increase in
photosensitivity, (i.e. a loss of the requirement of
darkness) with the result that the seeds become indifferent
to light. On the other hand, Wesson and Wareing (1969a,
1969b) describe experiments the results of which indicate
that a photorequirement is induced in the seeds of some
light-indifferent species during burial in the soil. The
expression of photorequirement in these experiments took the
form of a decline in the percentage of germination in darkness,
rather than an increase in percentage of germination in the
light.

# Reports of response to light in species of Amaranthus

In addition to foregoing detailed research into the effects of light on germination of the seeds of Amaranthus species, several authors have made general statements with respect to these species.

Rojas-Garciduenas and Kommedahl (1960) reported that there was a significant reduction in germination of seeds of Amaranthus retroflexus in continuous light as compared with either continuous darkness or alternating light and darkness. Crocker (1916) stated that this species has light-inhibited seeds. Barton (1945)wrote, "Tests have indicated that light does not favour germination, so could be eliminated The quality of reasoning revealed in this sentence appears to match the quality of English in which it Evans (1922) stated that the inhibition of was written. germination in light occurs even when the seed coats are "treated". In contrast to these reports for A. retroflexus, seeds of  $\underline{A}$ . hybridus are said to be dependent on light for germination (Engelhardt, Vincente and Silberschmidt, 1962). 5.5.4 Moisture as a factor controlling germination

The metabolic activity of a dry seed is remarkably low, probably as a direct result of its very low water content (Mayer and Poljakoff-Mayber, 1963). Reserve materials are stored in the seed in dehydrated form and growth can only resume after their hydrolysis. Thus sufficient water must be present before germination can occur. Also the dry seed is capable of withstanding drought conditions that would

prove fatal to a developing seedling and the stimulatory effect of water on germination ensures that the seedling will emerge into an environment with sufficient moisture. Species vary in the range of moisture tensions within which the germination of their seeds can occur. Seeds of some species have wide ranges, while seeds of other species have very specific requirements (Mayer and Poljakoff-Mayber, 1963).

Germaintion may be inhibited when the seeds are inundated with water, but in some species this condition may stimulate germination. The amount of oxygen dissolved in the water may condition this response.

The impermeability of seed coats to water provides a mechanism for maintaining dormancy. Loss of dormancy may arise naturally from damage to the seed coat by mechanical abrasion, microbial attack, passage through the digestive tract of animals or exposure to alternating high and low temperatures which by expanding and contracting the seed coat, will crack it.

Weise and Davis (1967) compared the germination of three species, including Amaranthus retroflexus, under two soil moisture regimes (for other details of this experiment see page 185). The different moisture regimes were obtained by (a) daily watering, and (b) watering for three days after sowing and then allowing the soil to dry. At alternating temperatures of 35/50°F and 50/65°F emergence was greater under the wetter conditions, while at temperatures of 65/80°F and 80/95°F germination was fast and emergence

occurred in the dry treatment during the three-day wet period. Consequently there were no differences in emergence between moisture treatments at higher temperatures.

Moisture conditions during dormancy may also influence the after-ripening process. As Mayer and Poljakoff-Mayber (1963) point out:

"After-ripening often occurs during dry storage. In other cases storage of the seeds in the dry state does not cause after-ripening. The seeds must be stored in the imbibed state, usually at low temperatures, in order that they after-ripen."

Lubke and Cavers (1969) were able to simulate the natural after-ripening of seeds of <u>Saponaria officinalis</u>.

In laboratory experiments they determined that soaking was more important than low temperatures in overcoming a dormancy imposed by the seed coat.

As discussed on page 181, Barton (1945) reported that seeds of Amaranthus retroflexus could germinate at constant temperatures as low as 20°C after two or three months of dry-storage, while moist-stored seeds were unable to germinate at this temperature after two years of storage. Barton (1945) examined the effects of removing seeds from moist storage at 20°C, allowing them to dry for different periods of time and then setting them to germinate at 20°C. With as little as three hours drying, 61% germination was obtained, compared with none in the non-dried control.

The behaviour observed by Barton may have implications for the control of germination under natural conditions.

Seeds buried in the soil may experience similar conditions

to seeds stored in continuous moist conditions. Agricultural or natural disturbance may expose the seeds allowing them to dry out and also placing them in a more suitable position for subsequent germination. With the first return of moist conditions and suitable temperatures, the seeds on the soil surface may then germinate.

5.5.5 Control of germination by gaseous substances

Mayer and Poljakoff-Mayber (1963) reviewed the influence on germination of variations in the composition of The availability of oxygen in the ambient the atmosphere. environment is an important factor in controlling germination. This control is achieved since germination involves respiration which is usually dependent on oxygen. In most species, the percentage of germination of seeds is proportional However, there to oxygen concentration within broad limits. are a few species in which a greater proportion of seeds can germinate at sub-normal oxygen concentrations. High carbon dioxide concentrations often inhibit germination but at the same time enhance the maintenance of viability in dormant Lipp and Ballard (1959) have shown that an atmosphere seeds. containing 2.5% carbon dioxide stimulates very high percentages of germination in dormant seeds of a range of leguminous species.

Seeds of Amaranthus retroflexus have been shown to germinate well in atmospheres (a) with as little as 10% oxygen, or (b) with as much as 15% carbon dioxide in the presence of 20% oxygen (McWilliams, 1966).

Ketring and Morgan (1969) have examined the role of ethylene in the germination of seeds of Virginia-type peanuts. They found that ethylene is given off by germinating seeds. Ethylene has also the effect of inhibiting the germination of seeds of the same species that have not germinated. This provides a regulatory mechanism that might lead to a continuous or intermittent sequence of germination of the seeds.

5.5.6 Other external agents that control germination

There are other agents in the external environment of a seed that may affect germination. Excluded from consideration here are those substances that are known only to bring about the death of the seed.

Abdul-Wahab and Rice (1967) have examined the inhibitory effect of Sorghum halepense on the germination of seeds of several species, including Amaranthus retroflexus, with which it is associated in abandoned fields. In experiments in petri-dishes, extracts of living leaf and rhizome inhibited both germination of the seeds and growth of the seedlings of A. retroflexus. Germination trials were conducted also in soils in which either leaves or rhizomes of Sorghum had been allowed to decay. Germination was inhibited in soil containing rhizome remains but was unaffected in soil containing leaf remains.

Gregg and McCormick (1966) suggested that the poor growth of several species, including Amaranthus hybridus, in uncultivated plots in which they had been grown in the previous season, involved self-inhibition.

Man may introduce substances into the environment which incidentally influence the germination of seeds. For example, seeds of Amaranthus retroflexus have been found to respond to 2,4-D, a foliar-contact herbicide which is toxic to many broad-leaved weeds (Rojas-Garciduenas and Kommedahl, 1960). Three-month old seeds of this species that had been scarified with sulphuric acid gave slightly greater germination after short exposures to all of the concentrations of 2,4-D examined than seeds that received no exposure to 2,4-D (Table 5.3). However, the germination of scarified seeds was strongly inhibited by exposures of five hours or more to all concentrations of 2,4-D. germination of non-scarified seeds was stimulated by all contrations with up to five hours exposure but inhibited with higher concentrations at exposures of 20 hours or longer.

Potassium nitrate has been found to stimulate the germination of seeds of Amaranthus hybridus (Engelhart, Wincente and Silberschmidt, 1962). The percentage of germination that resulted from two hours illumination given during a period of drak imbibition depended upon the time at which the illumination was given. Seeds immersed in 0.2% potassium nitrate produced greater germination than seeds immersed in water following illuminations given at any time throughout the period of darkness.

TABLE 5.3

GERMINATION OF SCARIFIED AND NON-SCARIFIED SEEDS OF AMARANTHUS RETROFLEXUS FOLLOWING IMMERSION IN DIFFERENT CONCENTRATIONS OF 2,4-D FOR DIFFERENT PERIODS OF TIME (Modified from Rojas-Garciduenas and Kommedahl, 1960 tables 1 and 2)

	immersion	Seeds*	imme	ent gent rsion in owing of	in 2.4	-D at '	tne
2,4-D	Water	ರ್ಲಿಕರಣ	0	5	25	50	500
0	30	s Ns	41 42				
1	29	S NS		48 45	46 68	50 68	46 48
5	25	s NS		3 60	3 60	1 70	1 46
20	10	s ns		4 68	1 58	2 47	1 33
30	0	s ns		3 65	3 55	0 36	0 29

\*Note: S = seeds scarified with H<sub>2</sub>SO<sub>4</sub>

NS = non-scarified seeds

# 5.5.7 Germination in different soils

Differences between environments in the field are rarely the result of variations in one factor. On the contrary, each environment is a complex system involving the interaction of many suspected and unsuspected variables; neighbouring environments may differ from one another in many ways. For example, soils that are classified as different types may differ in many variables of importance to seed germination. A few of these variables can be listed as examples: the capacity to absorb and retain heat,

retain moisture, and the composition of the soil solution. It would be unrealistic to try to control each of these variables experimentally while investigating the germination response of seeds. By comparing responses between seeds on different soil types, those combinations of conditions that occur most frequently in the field are included. In following this latter approach it is difficult to determine causal factors and the mechanisms whereby they act, but in experiments performed with care, the results are likely to be of greater relevance to the natural situation than the results of investigations of single factor effects.

Wiese and Davis (1967) examined the germination of seeds of three species, including Amaranthus retroflexus, in soils of two different types. At the highest temperatue regime, greater germination occurred on Amarillo fine sandy loam than on Pullman silty clay loam. A possible reason for this difference was that after daily watering the surface of the Amarillo soil remained wet longer than the surface of the Pullman soil.

Cuapinera and Guerrero (1968) examined the germination responses of seeds of Amaranthus hybridus in sterilised and non-sterilised soils of three types; sand, loam and clay. For each soil type, germination was greater in non-sterilised than in sterilised soil and they attributed this to the influence of micro-organisms in the non-sterilised soil. Differences were observed in the germination response in

different soils, but these revealed no trend, varying with the depth at which the seeds were buried and the age of the seeds (fresh versus five-year stored).

# 5.6 Genetic control of germination

Ultimately all germination responses are controlled by the genotypes of the parent plant and the seed embryo, since it is these that determine the absolute range of environmental conditions to which a seed can respond by germinating. The genotypes also determine the metabolic pathways on which environmental variables can act to influence germination.

Several authors have reported differences in the germination responses of seeds of one species collected from different parts of the same plant, from different plants in the same site and from plants in different sites (Barton, 1945, Cook, 1962, Thurston, 1962 and 1963, Cavers, 1963 and McWilliams, 1966). Cavers and Harper (1966) commented upon the lack of agreement among the results obtained by different workers investigating the effects of the "same" treatment in one species, Rumex crispus. They concluded:

"The difference in the behaviour of seeds in the hands of different observers may be due to any of the following factors: (a) Difference in the experimental techniques; (b) Difference between the behaviour of geographically distinct or ecologically distinct populations of the same species; (c) variations in the behaviour of seeds from individual plants of the same species; (d) variations in the behaviour of seeds from different parts of the inflorescence; and (e) variations in the ripeness of seed and conditions of storage before testing.

Very often the techniques used in germination tests are very often the techniques used in germination tests are incompletely reported and it is difficult to determine how much disparity is due to (a) above."

Harper (1965) noted that weed researchers in different parts of the world often describe quite different germination requirements for seeds of the same species. He suggested that the disparities "may well reflect evolutionary divergence in germination requirements". He doubted the widely held assumption that a species, population or even the progeny of one individual have in common a characteristic requirement for germination.

Experimental evidence of the inheritance of dormancy has been shown by Morley (1958) in <u>Trifolium subterraneum</u>. In seeds of this species, the genotype of the embryo determined, at least in part, the expression of dormancy by the seed. The genotype of the testa did not determine seed dormancy.

McWilliams, Landers and Mahlstede (1968) compared the weight and germination response of seeds produced by plants of Amaranthus retroflexus grown under uniform conditions from seeds collected over a wide geographical range. They discovered clinal variations in germination at 20°C and in seed weight that were associated with differences in latitude of the original collection. Seeds from northern collections were the heaviest and gave the greatest percentage of germination at 20°C. Although these plants were grown under uniform field conditions, differences in the photoperiodic response of plants from different collections meant that the plants flowered and set seed during different periods. Plants from northern collections flowered first

and further experiments were performed to determine whether the differences in seed weight and germination were a result of the photoperiodic response.

In one of these experiments, plants from different collections were grown under very short daylengths causing them to flower simultaneously. The weights and germination responses of seeds produced by these plants differed between collections as they had when the plants were grown under natural daylengths.

Weedy species often exhibit a polymorphism among the seeds from individual plants (Harper 1965). Seeds may be of visibly different types (Milliams, 1962, Robocker et al, 1969) or differences in their physiology may be inferred from their behaviour in germination experiments (Cavers and Williams (1962) found four different types Harper, 1966). of seed on individual plants of Chenopodium album; black seeds or brown seeds, each of which may be either reticulate -Brown seeds had no special requirements or smooth-surfaced. for germination, but smooth-black and reticulate-black each required specific treatments to produce complete germination in a sample. The frequency with which the smooth and reticulate types of seeds occurred upon a plant seemed to follow a cline in the British Isles from southeast to northwest.

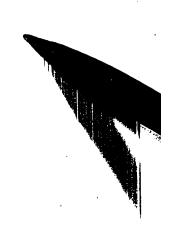
Steiner (1968) has demonstrated that much variation exists in the germination response of individual seeds of Oenothera even although the seeds are known to be of an

identical genotype. He described this phenomenom as "germination polyphenism" and suggested that differential pre-conditioning may explain its occurrence.

# 5.7 Summary

There is abundant evidence of the influence of environmental variables upon the dormancy and germination of seeds. Relatively little attention has been given to the effects of environmental pre-conditioning on subsequent germination behaviour although this appears to be an important factor in some species. Many authors have investigated the relationships between seeds and their environment during the period from maturation until germination. Much valuable information has been accumulated from these studies but in many instances observations made in laboratory studies have not been related to the behaviour of seeds under natural conditions.

The germination behaviour of seeds of A. retroflexus has received the attention of many authors but studies of the behaviour of seeds of A. hybridus and A. powellii are few and leave many aspects of the behaviour of seeds of these species uninvestigated.



## CHAPTER 6

# PRELIMINARY GERMINATION EXPERIMENTS

# 6.1 Introduction

Eight preliminary germination experiments were designed to obtain information from which more critical investigations could be performed. Various environmental factors, such as light and temperature were manipulated in these experiments in order to provide information on the conditions under which germination would occur. Other experiments were designed to determine whether handling the seeds for experimental purposes introduced unsuspected variation in germination response.

The factors examined in each of the eight experiments are listed in table 6.1. In experiments 1 to 3 seeds of several collections of <u>A. powellii</u> and <u>A. retroflexus</u> were set to germinate in alternating light and darkness under different temperature regimes and after different lengths of storage between harvest and germination. Different collections of each species were included to give a measure of the amount of variation within a species.

While preparing seeds for experiments 1 to 3 it was noticed that the samples of seeds were not homogeneous.

.;

·			
Experiment number	1	2	3
Factors examined	species differences	collection differences	collect
Species and collections included	P1,R1 (+B1, B6,T1)	R1,R2	P1,P2,F P5,P6,F R3,R4,F
Date of seed collection	see table 6.2	see table 6.2	see tab
Date of beginning of experiment	23-9-66	5-10-66	22-11-6
Storage conditions before the experiment2	seed room	seed room	seed ro
Germination trial:			
Incubator, growth room <sup>2</sup>	13/1	13/1	370
Thermoperiod (hours at maximum temperature:hours at minimum temperature)	12:12	12:12	16:8
Maximum temperature	25°0	25°C	30°C
Minimum temperature	10°0	10°C	18°0
Photoperiod (hours of light/hours of darkness)	12/12	12/12	16/8
Number of replicates/treatment	4	4	4
Number of seeds/replicate	100	100	75
Record of germination	daily for 35 days, weekly for a further 1½ months	daily for 24 days, weekly for a further 1½ months	daily f days, v for a f 2% mont
Results in table: <sup>3</sup>	A3.1	A3.2	A3.3
Illustration of results	Fig. 6.1	Fig. 6.2	Fig. 6.
Notes: 1 - Details of species a table 6.2.		·	2 – Add and

TABLE 6.1 A SYNOPSIS OF THE DETAILS OF PRELIMINARY EXPERIMENTS 1 to 8

3	4	5	6	7			
collection differences	seed differences	species and collection differences	experimental techniques, light regimes, species and collection differences	win con			
P1,P2,P3,P4, P5,P6,R1,R2, R3,R4,R5	R5	P2,P3,R1,R5	P2,P3,R1,R5	P5			
see table 6.2	see table 6.2	see table 6.2	see table 6.2	29-			
22-11-66	12-12-66	10-12-66	25–1–67	25-			
seed room	seed room	seed room	seed room	see			
37C	370	13/2	37C	37C			
16:8	16:8	24:0	16:8	16:			
30°C	30°0	30°C	30°C	30°			
18°0	18 <b>°</b> C		18°C	18°			
16/8	16/8	0/24	16/8 or 0/24	16/			
	t.	4	4	6			
4	4						
75	25	100	100	50			
daily for 18 days, weekly for a further 2½ months	daily for 4 days, alternate days for 8 days, weekly for 2 weeks	once after 7 days	daily for 7 days, weekly for 33 days, treatments 3 and 4; first time on day 7	dai			
A3.3	A3.4	6.6	A3.5	6.7			
Fig. 6.3	Fig. 6.4		Figs. 6.5 to 6.8				
2 - Additional details of conditions in the seed room, growth rooms 3 - and incubators are given in table 6.3.							

# NARY EXPERIMENTS 1 to 8

••		
6	7	8
experimental techniques, light regimes, species and collection differences	wintering conditions	storage conditions
P2,P3,R1,R5	P5	R1
see table 6.2	29-9-66 & 24-5-67	18–1–68
25 <b>-</b> 1 <b>-</b> 67	25-5-67	18-1-68
seed room	seed room or none	none
37C	37C	a)37C b)37/1
16:8	16:8	a)24:0 b)24:0
30°C	30°C	a)25°C b)35°C
18°C	18°C	
16/8 or 0/24	16/8	a)12/12 b)16/8
4	6	6
100	50	50
daily for 7 days, weekly for 33 days, treatments 3 and 4; first time on day 7	daily for 7 days	daily for 9 days
A3.5	6.7	A3.6
Figs. 6.5 to 6.8		Figs. 6.9 and 6.10
d room, growth rooms	3 - Tables beginning other tables as	ng A3 are in Appendix 3 re in this chapter.

Most seeds appeared almost black but a small number appeared chestnut-brown. It was suspected that these seeds had been immature when harvested but, alternatively, they might have represented a different kind of seed with perhaps a different germination response. Other seeds were observed that had been partially eaten by larvae of the case bearing moth, Coleophora lineapulvella Chamb. Another group of seeds were conspicuous by their very small size. Experiment 4 was designed to determine (1) if these different kinds of seeds differed in their germination behaviour, and (2) whether any of them should be excluded from further experiments.

Experiments 1 to 4 were conducted under conditions of alternating light and darkness, thus simulating conditions that might naturally be encountered by seeds on the soil surface. Buried seeds occupy a different environment from seeds on the surface of the soil and one of the ways in which the environment of buried seeds differs is in the absence of light. Experiments 5 and 6 were designed to compare the germination response of seeds in darkness with that in alternating light and darkness.

The seeds used in experiments 1 to 6 had been stored in the seed room for different lengths of time before they were used. Moreover none of these experiments were conducted at the time at which seeds naturally germinate in the field. Experiment 7 was designed to compare the germination response of seeds stored in the seed from throughout the winter and

those that had passed the winter in a natural situation.

The experiment was conducted at a time at which seedlings of the three Amaranthus species were appearing in the field.

Barton (1945) reported that seeds stored in dry conditions for two or three months were able to germinate over a wider range of temperatures than freshly harvested seeds. Experiment 8 was designed to determine how rapidly changes of this nature occur. Such knowledge was essential in order that handling seeds for experimental purposes should not introduce additional factors to influence germination responses.

6.2 Materials and methods

# Experimental design

The different treatments employed to investigate each factor are given in table 6.1. In each experiment the replicates of each treatment were maintained under uniform conditions whenever possible. When the experimental conditions were not uniform (e.g. when two different shelves of an incubator were used) replicates of each treatment were assigned to randomised blocks to ensure that experimental errors were randomly distributed among the treatments.

# Materials

The collections from which seeds were used varied between experiments; they are listed in table 6.1. In this table collections are referred to by their code symbols (e.g. P5, R1 etc.) and an interpretation of these symbols together with information concerning the locality in which the collection

was made, the time of harvest and the existence of herbarium specimens are presented in table 6.2.

# Preparation of seeds

When collections of seeds were made in the autumn of 1966, substantial parts of the aerial portions of the plants were harvested, brought back to the laborator and allowed to dry in the air. The terminal and lateral inflorescences were removed from the plant remains and broken up by hand. Then the resulting mixture of seeds and chaff was shaken through a series of sieves in order to separate the seeds. The seeds that were cleaned in this manner were placed in open bags and stored in steel cabinets in the "seed room", a room in which both the temperature and humidity were under close control (for conditions in this room see table 6.3). Seeds remained in the seed room until they were used in an experiment.

In experiment 7 one of the treatments consisted of seeds collected from plant remains after the winter had passed. The preparation of these seeds was identical to that described above. However, the seeds were transferred directly to experimental conditions after cleaning.

The manner in which the seeds used in experiment 8 were prepared differed from that described above. Seeds were collected from plants growing in the greenhouse without removing the inflorescences from the plants or uprooting the plants. The terminal and lateral inflorescences were gently shaken over a large sheet of paper. This caused any

TABLE 6.2

DETAILS OF THE SOURCES OF THE MATERIAL USED IN GERMINATION EXPERIMENTS

NUMBERS OF VOUCHER SPECIMENS*	23,53–57	77 78 77 77	10-0/60-60-	18,19,81–85	72-76,87	20,77-80	25,62–66	89,67-71		21,180-182		! !	177–179	183–185	(continued)
HABITAT V	Bean field			Bean field	Turnip field	Roadside	Waste ground	Bean field		Fallow field 21,180-182	Stable yard	Turnip field	Roadside	Potato field 185-185	
ION - REFERENCE*	264915	( ( (	08426/	244934	608014	304860	777598	592128		252035	304847	608014	697566	828645	
OF COLLECTION- COUNTY* REFE	Kent		of London Middlesex	Kent	Huron	Kent	Middlesex	Huron		Kent	Kent	Huron	Middlesex	Middlesex	
LOCALITY TOWNSHIP	Howard		City of London	Howard	Usborn	Howard	City of London	Tuckersmith		Howard	Howard	Usborn	Delaware	London	
DATE OF COLLECTION	18-9-66		15-9-66	18-9-66	7-10-66	19-9-66	29-9-66	7-10-66		99-6-8	18-9-66	7-10-66	6-10-66	20-10-66	
SPECIES AND COLLECTION NO.	A. hybridus	A. powellii	Ы	ъ2	P3	7 A	P5	P6	A. retroflexus	R	R2	R3	R4	85	ì



NUMBERS OF VOUCHER SPECIMENS*		ı		!	12,17	
NO VOICE OF THE PERSON OF THE					72	
навітат	,	Waste ground	Fallow field		River bank	
rion Reference*		778615	252035		792580	
LOCALITY OF COLLECTION INSHIP COUNTY* REF		of London Middlesex	Kent		of London Middlesex	
LOCALITY TOWNSHIP		City of London	Howard		City of London	
DATE OF COLLECTION		17-9-66	99-6-8		15-9-66	
SPECIES AND COLLECTION NO.	A. blitoides	B1	B6	A. tuberculatus	17	

- All counties are in the Province of Ontario. 7 \* Notes:
- References give position on the Universal Transverse Mercator Grid to the nearest 100 metres. 6
  - Voucher numbers are the author numbers (R.A.Frost) of specimens deposited in the Herbarium of the University of Western Ontario.

3



TABLE 6.3

# DETAILS OF ENVIRONMENTAL CONDITIONS IN INCUBATORS, GROWTH ROOMS AND THE GREENHOUSE

Environment	Light Quality H	condit eight I ft.	ions <sup>1</sup> ntensity f.c.	Tempera Day N:	ture <sup>2</sup>	Relative <sup>3</sup> humidity
Incubators:						
13/1	CML	1	300			NC
13/2	No il	luminat	ion			NC
13/3	CWF	1.3	1250 ) 800 )			75–95
16/1	CWF	1	300			NC
37/1	F	1	200			NC
Growth Rooms	:					
37C	CWF	5	1300			50 <b>–7</b> 0
37D	CWF	3	700			60-80
Greenhouse 1	9 D + CWF	3	1250m	25(14)	15(10)	NC
Seed Room	Light-tig	ht cabi	nets	21-2	5	40M
Cold Room	Light-tig	ht cani	sters	5		80m

Notes: 1 - Light intensities are average values measured at the distance indicated (height) from the source.

The quality of the light source is indicated by the following abbreviations:

CWF Cold white fluorescent, F Fluorescent, D Daylight.

<sup>2</sup> Temperatures are given only when these can not be varied. Figures in parentheses indicate the length of the thermoperiod in hours.

<sup>3</sup> Abbreviations are: NC no control, M maximum value, m minimum value.

mature seeds to fall from the inflorescences. The seeds were then removed from within the utricles (if these were still attached) by rolling the fruits between two sheets of bristol board. The seeds were assigned directly to experimental treatments.

# Pre-germination treatments

Some of the treatments included in experiment 8 consisted of different storage conditions before germination was examined. The details of these treatments are given in table 6.4.

TABLE 6.4

# DETAILS OF SPECIAL TREATMENTS EMPLOYED IN SOME PRELIMINARY EXPERIMENTS.

Experiment	Nature of the treatments		Details of the treatments
4	Four different seed fractions	1)	Black seeds of sound appearance and average size (mean weight 10.1 mg/25 seeds).
		2)	Black seeds of sound appearance yet abnormally small (mean weight 5.7 mg/25 seeds).
		3)	Black seeds showing partial insect damage.
		4)	Brown seeds of sound appearance and average size.
6	Four light regime combinations	1)	Alternating light and darkness for 40 days (scored daily for first seven days).

# Experiment Nature of the treatments

Details of the treatments

- 2) Continuous darkness for 40 days (scored daily for first seven days).
- 3) Alternating light and darkness for seven days followed by continuous darkness for 33 days (first scored on seventh day).
- 4) Continuous darkness for seven days followed by alternating light and darkness for 33 days (first scored on the seventh day).

### Four different 1) No storage. 8 storage treatments

- 2) Storage (in petri-dishes) for eight days, dry, in seed room\* before germination.
- 3) Storage (in petri-dishes) for four weeks, dry, in seed room\* before germination.
- 4) Storage (in petri-dishes) for four weeks, with 10 ml water, in cold room\* before germination.

\*Note: for details of conditions in seed room and cold room see table 6.3.

# Experimental conditions

Table 6.1 indicates the number of replicates of each treatment (or collection) in each experiment and the number of seeds included in each replicate. In experiments 1 to 7 the seeds were counted and placed on Whatman 0.4 mm germination paper in glass petri-dishes of 90 mm diameter and 10 mm depth. In experiment 8 the seeds were placed on Green's 450 filter

paper in petri-dishes.

On the day that each experiment was started 10 ml of distilled water were added to each petri-dish and the petri-dishes were arranged either at random, or at random within blocks, in the incubator or the growth room. Table 6.1 lists the dates on which the germination trials were begun, the identity of the incubator or growth room in which the experiment was conducted, and the major environmental conditions that were maintained during the experiment. Further details of the conditions in each growth room or incubator can be found in table 6.3.

In experiments 3, 4, 6 and 7, the petri-dishes were first arranged randomly in fours or fives in plastic dishpans, of approximate dimensions 30 cm by 36 cm by 10 cm deep. A piece of transparent polythene was secured over each dishpan in an attempt to reduce the rate at which moisture would be lost. Then the dishpans were arranged at random on the tables in growth room 37C.

In experiments 5 and 6 the "continuous darkness" treatment was achieved by arranging the petri-dishes at random in vertical columns in aluminum canisters 25 cm in height. The canisters were closed with light-tight lids and sealed with "Masking Tape" (made by 3M) to ensure that darkness was maintained.

# Recording germination

A seed was considered to have germinated when its radicle had emerged and developed a red pigmentation. At

this point its germination was recorded and it was removed from the petri-dish.

In those experiments in which germination was recorded while the seeds were being maintained in continuous darkness, the canisters containing the petri-dishes were opened only in a dark-room. The petri-dishes were illuminated with green light, provided by an electric flashlight fitted with green filters, in order to score the number of seeds that had germinated.

The frequency with which germination was recorded in each experiment and the duration of each experiment are presented in table 6.1. Those seeds that remained ungerminated at the end of experiment 4 were broken open and if they contained a bright, white embryo they were recorded as viable. Viability was not estimated in any of the other experiments.

# 6.3 Results

The cumulative germination scores for the replicates of each collection or treatment are presented in Appendix 3 for each experiment. Table 6.1 indicates the table in the appendix in which the results for a particular experiment appear. Table 6.1 lists also the numbers of the figures in which the mean results for each treatment and collection are illustrated. Some of the day to day scores have been omitted in some of the figures in order to maintain clarity.

The estimates of viability for the different seed fractions in experiment 4 are presented in table 6.5.

# TABLE 6.5

# ESTIMATES OF THE VIABILITY OF SEEDS IN EACH FRACTION EXAMINED IN EXPERIMENT 4

# SEED FRACTION VIABILITY 1) Black seeds, sound appearance, average size 24.75 per 25 2) Black seeds, sound appearance, very small 24.50 per 25 3) Black seeds, damaged by insects 0.25 per 25 4) Brown seeds Values are the means of four replicates.

# TABLE 6.6

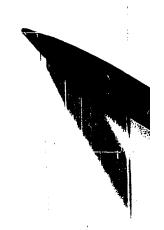
A KEY TO THE SYMBOLS USED TO DENOTE SPECIES AND COLLECTIONS IN THE FIGURES INCLUDED IN CHAPTERS 6 AND 7

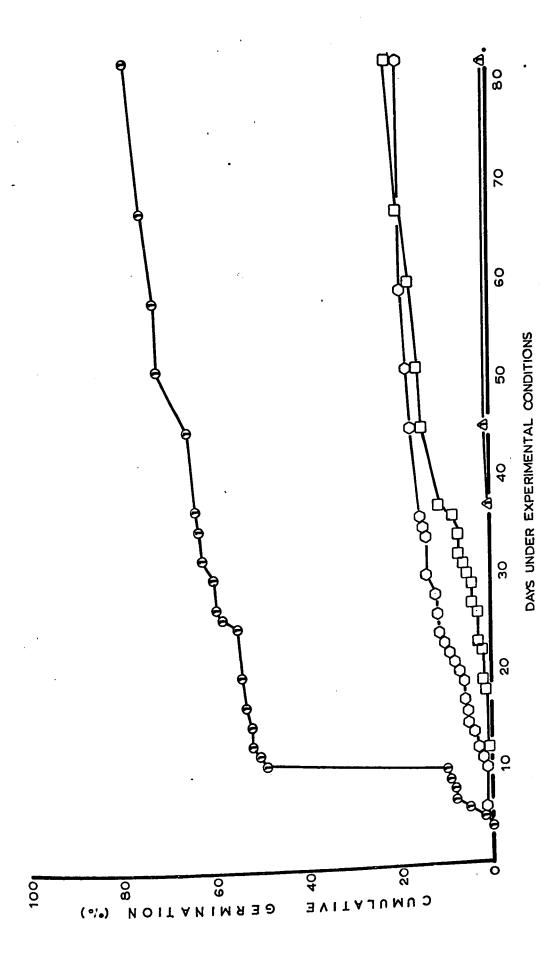
# a) Species:

- ☐ A. blitoides
- A. hybridus
- △ A. powellii
- O A. retroflexus
- O A. tuberculatus

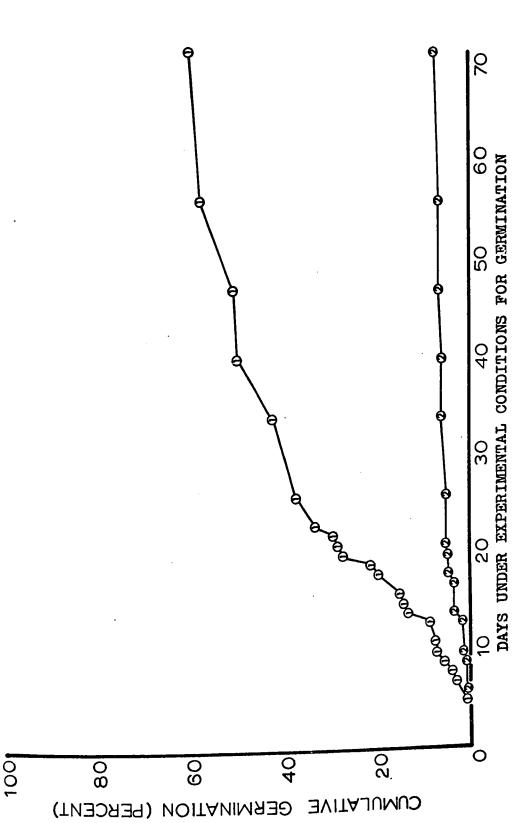
# b) Collections:

Numbers in each of the open symbols given above refer to the collections of each species. These numbers correspond to the numbers describing collections given in table 6.2.

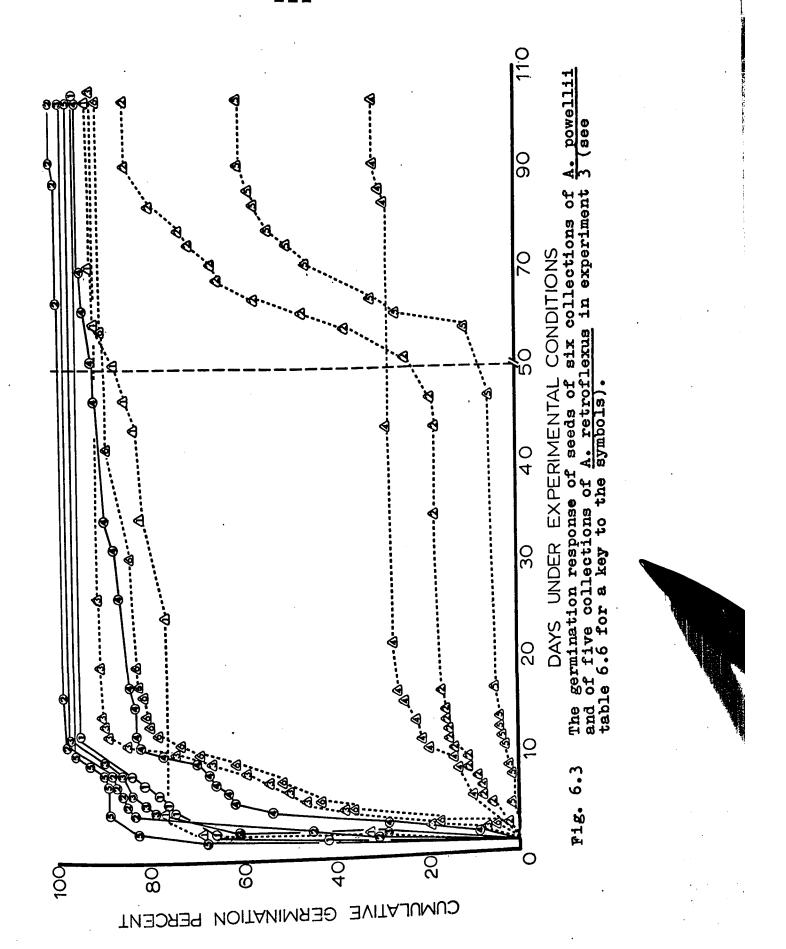




seeds of four different species of (see table 6.6 for key to symbols). The germination response of Amaranthus in experiment 1 F18. 6.1



The germination response of seeds of two different collections of A. retroflexus in experiment 2 (see table 6.6 for key to symbols) Fig. 6.2



## 6.4 Discussion of the results

## Experiments 1, 2 and 3

In examining the results of these three experiments attention can be focussed on three things: (1) Variation between the collections of one species; (2) Variation between the two species, <u>A. powellii</u> and <u>A. retroflexus</u>; and (3) Variation within one collection at different times and temperatures.

a substantial difference in germination response between seeds of collections R1 and R2 (Fig. 6.2). In experiment 3, however, seeds of these two collections were almost identical in their response (Fig. 6.3). These two experiments differed in two important respects. Firstly the alternating temperatures of experiment 2 were 25% and 10°C while for experiment 3 they were 30° and 18°C. Secondly, experiment 3 was conducted about 8 weeks after experiment 2, yet with seeds of the two collections in question collected at the same time. In the period between experiments the seeds had been stored in the unnatural environment of a storage room with controlled humidity and temperature.

Thus, although seeds of the different collections of A. retroflexus showed an almost uniform response in experiment 3, it can not be assumed that their responses would be the same under different conditions.

Variation between the collections of A. powellii was demonstrated in experiment 3 (Fig. 6.3). The collections of this species fell into two groups on the basis of the germination response of their seeds. In one group germination was immediate and high percentages of germination were rapidly achieved. In the other group there was little germination until after 50 days in the experimental conditions, and even then germination did not reach 100%. Thus with respect to the germination conditions of experiment 3, A. powellii can be seen to include collections that differed in their seed dormancy. Seeds of the first group could be said to be mostly non-dormant, while those of the second group were mostly dormant but showed signs of after-ripening under the experimental conditions.

- were exhibited in experiments 1 and 3. In experiment 1 collections R1 and P1 showed very different responses, with almost all seeds of collection P1 dormant (Fig. 6.1). In experiment 3, collections P1, P3 and P6 showed only small differences from all collections of A. retroflexus, in their rate of germination (Fig. 6.3). On the other hand, collections P2, P4 and P5 differed markedly from A. retroflexus (and from the other collections of A. powellii) in both the rate and ultimate percentage of germination of seeds.
- (3) Collection R1 was included in each of the three experiments, and the germination pattern of its seeds differed in each one. In experiment 1, when the seeds were

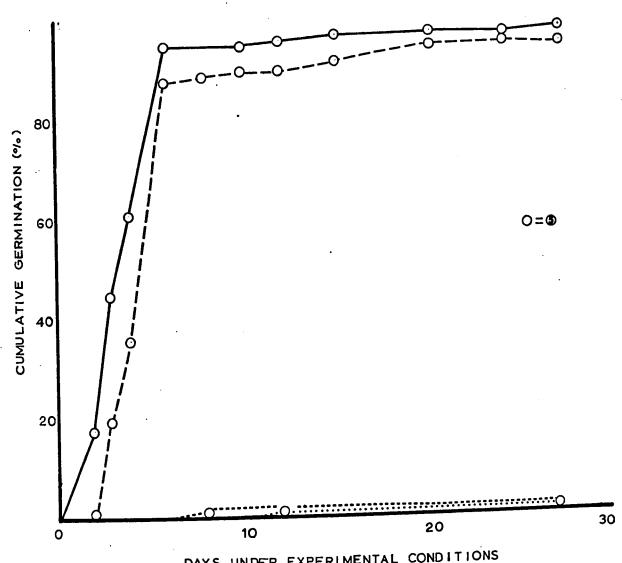
15 days old (i.e. 15 days after harvest) germination was fairly rapid and over 75% of the seeds had germinated in 80 days (Fig. 6.1). In experiment 2, which was conducted under identical conditions to experiment 1, the seeds were 27 days old when introduced to the experimental conditions. Germination was much slower (Fig. 6.2). After 70 days almost 60% of the seeds had germinated whereas this number had germinated in 25 days in experiment 1. In experiment 3, which was conducted at higher temperatures and about eight weeks after experiment 2, seeds of collection R1 germinated very rapidly; over 90% germinated in 15 days (Fig. 6.3).

Vegis (1964), cited A. retroflexus as an example of those plants whose seeds acquire a dormancy in which the temperature range that will permit germination narrows to The behaviour restrict germination to high temperatures. of seeds of collection R1 in these three experiments can be explained in this way. In Vegis's terms, the seeds were still in a stage of predormancy at the time of experiment 1. For a proportion of the seeds, the alternating 25°/10°C temperatures were within the range that permits germination, at the time of experiment 1. By the time of experiment 2 these temperatures were excluded as a result of a narrowing Experiment 3 was conducted at higher of the range. temperatures and with seeds that had been dry-stored for two It is possible that the temperatures and a half months. employed in experiment 3 were within the partial-dormancy

range. Alternatively, the seeds may have begun to afterripen by this time. Barton (1945) stated that seeds stored in dry conditions for two to three months were capable of germinating at temperatures as low as 20°C.

Any attempt to attribute differences observed in these experiments to any particular phenomenom must be qualified by a consideration of all of the possible sources of variation in the seeds used. These can be listed as follows:

- a) From table 6.1 it can be seen that the seeds were collected over a period of nearly two months. Thus the period of time that the seeds were stored in the rather abnormal conditions of the seed room varied from collection to collection and also varied from one experiment to the next.
- b) There is little reason to expect that the plants chosen for seed collection were at an identical stage of maturity, and therefore differences in maturity might have been responsible for some of the behavioral differences observed.
- c) Table 6.1 gives also a very general description of the habitat in which the parent plants were found. It can be seen that the habitats varied very much between collections. Differences probably existed in nutrient and water supply, in competition with other species and with individuals of the same species, in exposure to herbicides and many other environmental factors.
- d) In addition to the factors already mentioned, there is the possibility that the differences observed reflect inherent



DAYS UNDER EXPERIMENTAL CONDITIONS
FOR GERMINATION

KEY	
<u>Plack seeds</u> , of sound appearance and (1) average size	
(2) abnormally small	
with evidence of attack by insect	*********
Brown seeds  of sound appearance	

Fig. 6.4 The germination of different fractions of seeds of A. retroflexus in experiment 4.

genotypic differences between the different collections.

It is obvious then that the differences between collections observed in these experiments could not be attributed to any one factor since the effects of individual factors could not be isolated. Further experiments were therefore planned to elucidate the roles of particular factors in controlling germination behaviour.

## Experiment 4

The results of the observations on viability (table 6.5) indicated that brown seeds and those damaged by insects should be excluded from further germination experiments. Abnormally small seeds began to germinate slightly later than seeds of average size. Once germination had begun, it proceded to conclusion as rapidly in both samples. The difference in germination response was slight, and the number of small seeds was low. Consequently it seemed inappropriate to consider small seeds independently.

## Experiments 5 and 6

There are several interesting features in the results of experiments 5 (table 6.7) and 6 (Figs. 6.5 to 6.8). These can be discussed under the following categories:

# a) Influence of scoring the experiment

Experiment six provided an opportunity to observe the germination response of seeds in petri-dishes and in canisters that were either; (a) opened daily for seven days in order to score germination, or (b) opened only on the seventh day. The percentage germination of seeds after

seven days was almost the same in treatments 2 and 4, and also in treatments 1 and 3, for each collection (see table 6.4 for a description of the treatments). This indicated that opening the petri-dishes and canisters did not influence the amount of germination recorded.

## b) Darkness and Alternating light

No more than 60% germination occurred in continuous darkness in three of the collections examined; P3, R1 and R5 (Figs. 6.5 to 6.7). In these collections, seeds maintained under alternating light and darkness completed their germination (i.e. over 90% germinated) in 20 days. However, seeds of collection P2 behaved differently (Fig. 6.8). Of those seeds maintained under alternating light and darkness, only 30% had germinated in 40 days, while 90% of the seeds kept in continuous darkness had germinated in two days.

# c) Germination after a change in conditions

The 40% (approximately) of seeds of collections P3, R1 and R5 that failed to germinate in continuous darkness, germinated rapidly following transfer to alternating light and darkness (Figs. 6.5 to 6.7). Conversely, of the seeds of these collections that had not germinated in the first seven days in alternating light and darkness, fewer germinated subsequently among those transferred to continuous darkness than among those that remained in alternating light and darkness.

The response of seeds of collection P2 was again different (Fig. 6.8). Of seeds kept in alternating light and darkness, about 20% germinated in seven days, with little further germination when maintained under these conditions. Seeds that were transferred to darkness after seven days gave very little additional germination also. This was in contrast to seeds placed immediately into continuous darkness which rapidly gave 90% germination. Imbibition under alternating light and darkness could be considered to have induced a secondary dormancy in seeds of this collection.

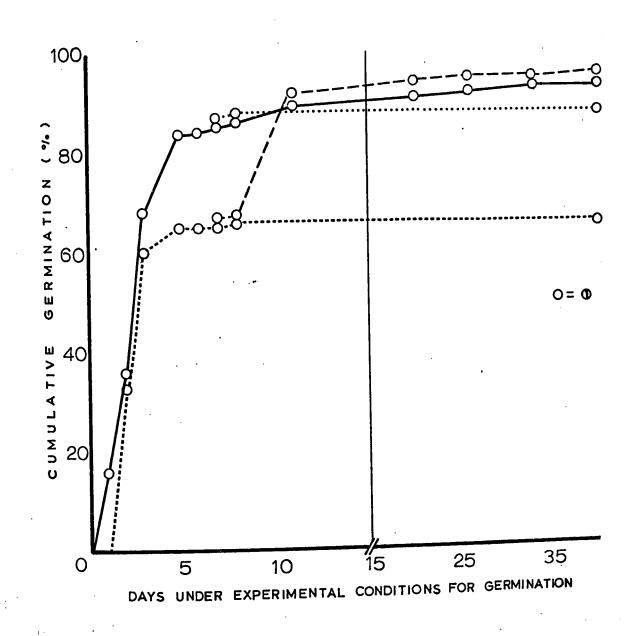
TABLE 6.7

TOTAL GERMINATION OF SEEDS AFTER SEVEN DAYS IN EXPERIMENT 5

Species:	A. retrof	lexus	A. powellii		
Collection:	R1	R5	P2	P3	
Total Germination:	94.5	78•5	67.3	43.8	
Totals, expressed a	s percenta	ges, are	the means	for 4	
replicates of 100 s	eeds each.				

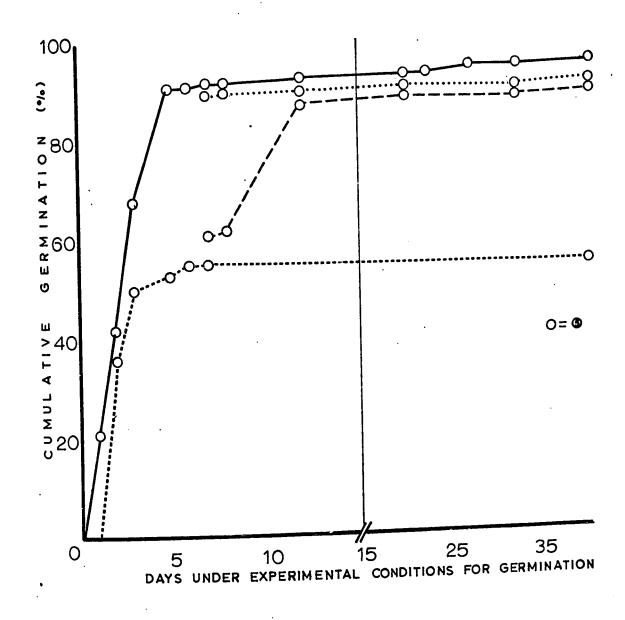
## d) Temperature differences

Experiment 5 (table 6.7) was conducted at a constant 30°C, whereas experiment 6 was conducted under alternating temperatures of 30° and 18°C. In other respects treatment 2 of experiment 6 is similar to experiment 5, and the two can logically be compared. In experiment 5 (constant temperature) collections of A. retroflexus gave greater



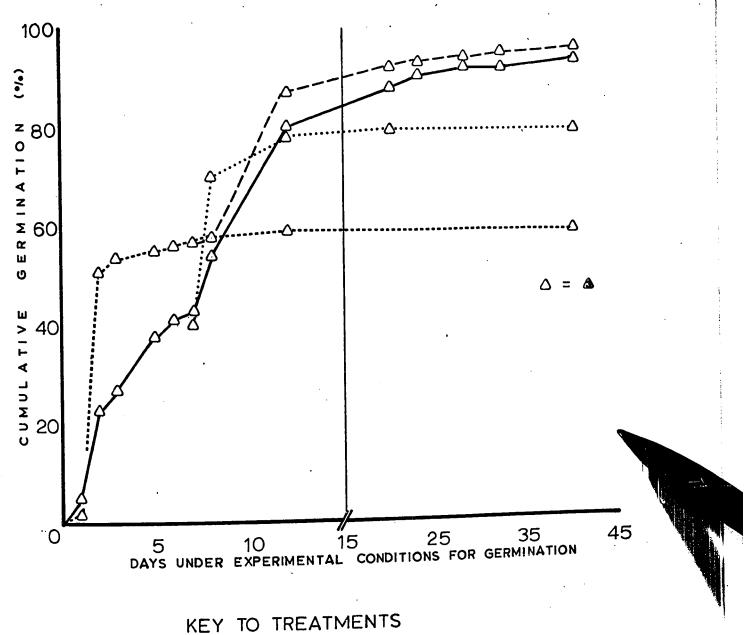
# KEY TO TREATMENTS

- Alternating light and darkness for the duration of the experiment
   Continuous darkness for the duration of the experiment
   Alternating and darkness for 7 days then continuous darkness
   Continuous darkness for 7 days then alternating and darkness
- Fig. 6.5 The germination response of seeds of A. retroflexus (collection R1) in experiment 6.



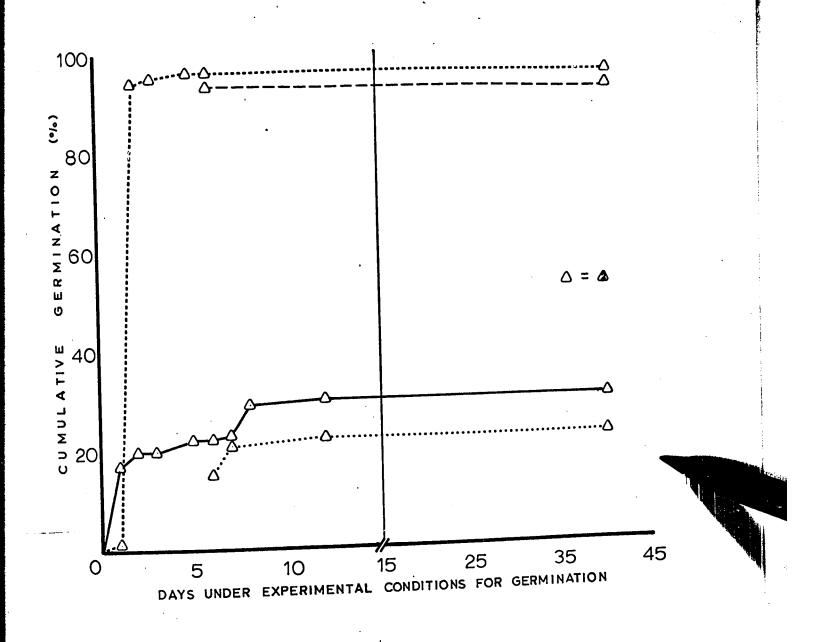
# KEY TO TREATMENTS

- Fig. 6.6 The germination response of seeds of A. retroflexus (collection R5) in experiment 6.



Alternating light and darkness for the duration of the experiment Continuous darkness for the duration of the experiment Alternating and darkness for 7days then continuous darkness Continuous darkness for 7 days then alternating and darkness

The germination response of seeds of A. powellii (collection P3) in experiment 6. Fig. 6.7



# KEY TO TREATMENTS

1. — Alternating light and darkness for the duration of the experiment
2. — Continuous darkness for the duration of the experiment
3. — Alternating and darkness for 7 days then continuous darkness
4. — Continuous darkness for 7 days then alternating and darkness

Fig. 6.8 The germination response of seeds of A. powellii (collection P2) in experiment 6.

germination in seven days than in experiment 6 (alternating temperatures). Collections of <u>A. powellii</u> behaved differently, giving greater germination in experiment 6 than in experiment 5.

## Experiment 7

#### TABLE 6.8

# TOTAL GERMINATION OF SEEDS AFTER SEVEN DAYS IN EXPERIMENT 7

Treatment:

Lab.-wintered

Field-wintered

Total germination:

14.7

49.7

Values are means of six replicates expressed as percentages.

The two samples of seeds used in this experiment were seen to behave differently in their germination response to the same conditions (table 6.8). Although the seeds had obviously encountered different conditions during the winter preceding the germination trial, other differences existed Firstly, although that may have influenced the results. the seeds were collected from plants in the same area (of size 50 x 50 ft. approximately), different plants were Thus the differences might reflect genotypic involved. Secondly, the seeds left on the plant during differences. the winter may not have been a representative sample of all the seeds produced by that plant. For this reason the seeds collected in the previous September may have differed in the proportions of seeds of various types from the seeds collected after the winter.

## Experiment 8

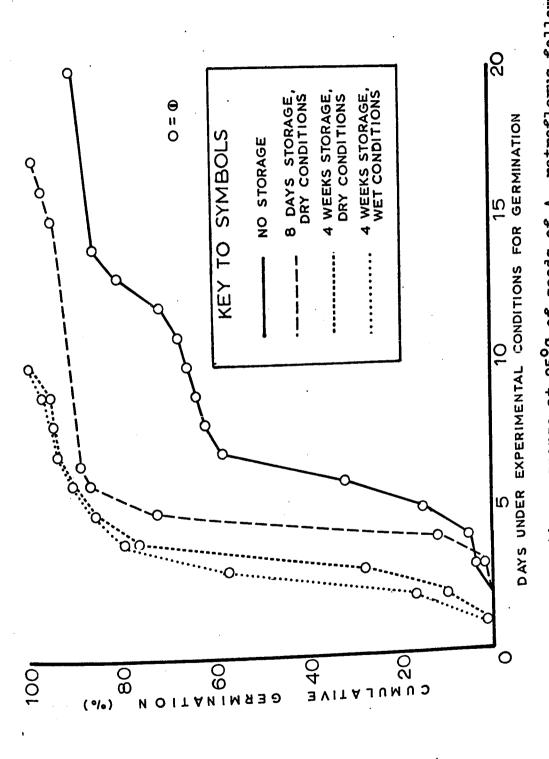
The response of seeds at 25°C was seen to be influenced by the length of storage and the conditions under which seeds were stored (Fig. 6.9). Thus, the longer seeds were stored the more rapidly they began to germinate and completed their germination. Seeds stored under moist conditions germinated more rapidly than those stored for the same length of time under dry conditions.

At 35°C the germination of seeds of each treatment occurred more rapidly and the differences between treatments were less pronounced (Fig. 6.10). At this termperature the chief difference was between seeds that received any storage and freshly harvested seeds. The former seeds were about one day ahead of the latter in their percentages of germination.

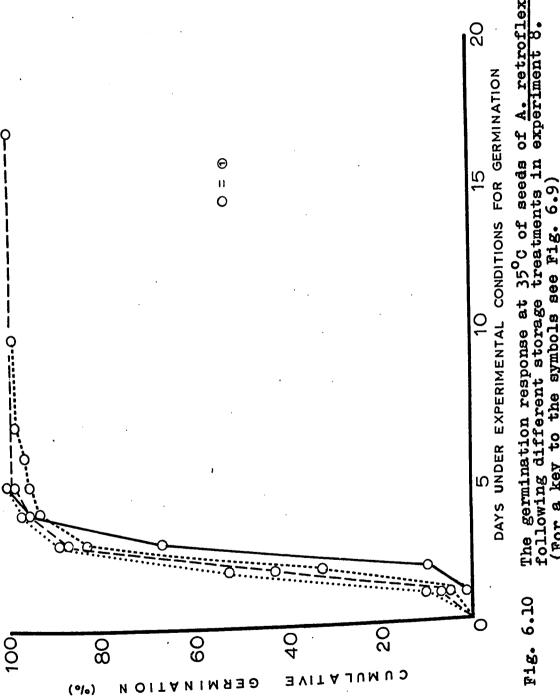
These results indicate that the storage of seeds is an important factor to consider in germination experiments.

When different treatments or collections are to be compared it is essential, if seeds must be stored, that they be stored under identical conditions for identical periods.

The results of this experiment contrast with the results of experiments 1 and 2. In the last mentioned experiments, it was suggested that seeds were undergoing pre-dormancy between days 15 and 27. In this experiment it appears that the seeds had begun to after-ripen before they were eight days old.



retroflexus following The germination response at  $25^{\circ}\mathrm{C}$  of seeds of A. different storage treatments in experiment 8. Fig. 6.9



(For a key to the symbols see Fig. 6.9)

#### CHAPTER 7

# AN EXPERIMENTAL EVALUATION OF SOME OF THE FACTORS INFLUENCING THE GERMINATION OF SEEDS

## 7.1 Introduction

It has been stated already that germination is a critical stage in the life histories of those species of plants in which reproduction is in some part dependent upon seeds. The nature of the germination response will determine in part the frequency and abundance with which a species occurs. Discontinuities in the distribution of a species may reflect the degree to which the germination response is appropriate in different environments. Thus the differences in the frequencies and distribution of the three species (reported in Chapter 4) may have resulted from differences in the germination responses of their seeds.

The results of preliminary experiment 3 demonstrated differences in germination responses between seeds of different collections of A. powellii and between seeds of some collections of A. powellii and A. retroflexus.

During the discussion of the results of experiment 3, some factors were proposed that might have influenced the germination responses that were observed. The following

experiments were designed to clarify the nature of these responses, to attempt to determine the causative factors and to evaluate, where possible, the ecological significance of the relationships.

Many of the studies (described in chapter 5) of the factors that influence germination have been concerned with the effects of isolated factors on freshly harvested or dry-stored seeds. While indicating that different treatments may bring forth different germination responses, these investigations do not prove that such factors are of importance under natural conditions. On the other hand, attempting to work with seeds under natural conditions proves difficult. Very few of the effects of individual factors can be isolated. It is impossible to control factors independently and even if such control were possible the potential number of combinations of conditions would be enormous.

An approach that may be rewarding is to manipulate the plant or the environment (or both) in such a way as to simulate the major alternatives in the conditions that may be experienced naturally by the plants or the seeds. In following this approach combinations of factors are varied. The results of experiments based on this approach may describe with accuracy the natural behaviour of the species but it is unlikely that they will isolate the effects of individual factors. However, the results of such experiments may provide the rationale for further investigations of the

influence of individual factors.

In most of the experiments to be described, the approach of manipulating plants and environments, outlined above, has been employed.

# 7.2 Factors examined for their role in controlling germination

Nine different "factors" were examined for their role in controlling the germination of seeds. Most of these "factors" were in fact complex interactions of two or more individual variables. In the following paragraphs a brief summary is given of the factors that were examined and table 7.1 indicates which factors were included in each experiment.

The first three factors included variables that might have influenced the seeds while they were on the parent plant. These factors were:

- 1) The time at which the seeds that gave rise to the parent plant were sown (subsequently described as "sowing time").
- 2) The time at which the seeds became mature ("harvest time").
- The position at which the seeds were produced on the parent plants ("position on plant").

The next factor included variables that might have influenced the seeds after maturity either on or off the parent plant. This factor was examined in general form:

4a) The conditions experienced by seeds between maturity

and the time of germination ("post-harvest treatments" or "wintering conditions").

More specific aspects of this factor were also examined:

- The position relative to the soil surface, in which the 4b) seeds wintered.
- The influence of the decomposing remains of the parent 4c) plant during wintering ("plant remains").

The next group of factors included variables that might have influenced the seeds at the time of germination:

- Differences between conditions in the field and 5) conditions in the laboratory.
- The influence of the fruit coat or utricle ("utricle"). 6)
- The influence of light. 7)
- The influence of temperatures. 8)
- The influence of soil conditions. 9)

In addition to these nine factors the experiments that follow were also designed to evaluate the influence of genetic differences in the control of germination.

TABLE 7.1 FACTORS EXAMINED IN GERMINATION EXPERIMENTS 9 TO 13

Experiment number	Factors examined
9	Sowing time, harvest time, post-harvest treatments, field versus laboratory, temperature, genetic differences.
10	Position on plant, field versus laboratory, utricle, light, temperature, genetic differences.
11	Soil conditions, genetic differences. (continued)

(continued)

# Experiment number

#### Factors examined

- 12 Field versus laboratory, genetic differences.
- Position relative to soil surface, plant remains, genetic differences.

## 7.3 Experiment 9

This experiment was designed to examine the effects of harvest and sowing times on preconditioning as expressed in Since preconditioning the germination behaviour of the seeds. effects may be subsequently modified by conditions between the time of seed maturity and natural germination, the effects of different post-harvest treatments were also The number of different collections that investigated. could be included was limited by the number of treatment Three collections of  $\underline{A.}$  powellii were used combinations. as experiments 3 and 6 had shown distinct variation within The previous response of collections of this species. A. retroflexus had been almost uniform and only one collection was used in this experiment. Only one collection of seeds of  $\underline{\text{A. hybridus}}$  was used since it was the only one available at the time this experiment was initiated.

## 7.3.1 Materials and methods

# 7.3.1.1 Culture of plants under uniform conditions:

Plants were grown from seed of the following collections: H7, P1, P2, P3, R1. Details of the habitats from which these collections were made are presented in table 6.2 page 208.

At approximately monthly intervals, seeds of each collection were set to germinate in petri-dishes. days after sowing, seedlings were transferred to peat pots containing a standard potting soil. The seedlings were maintained in pots in the greenhouse for between three and Then, 25 plants of each collection were four weeks. planted one metre apart in rows which were also one metre apart in Guelph loam soil at the Department of Botany Experimental Farm. A synopsis of the dates of sowing and transplanting for each sowing time is given in table 7.2.

TABLE 7.2 DETAILS OF THE DATES OF SOWING AND TRANSPLANTING FOR EACH SOWING TIME

Sowing time	1	2	3	4
Date on which	10/5/67	24/5/67	28/6/67	28/7/67
Date on which plants were transferred to the field	1/6/67	20/6/67	24/7/67	20/8/67

# I take a feet of

## 7.3.1.2. Seed harvests

Harvests were planned at different times\*to allow comparison of seeds from different harvests of plants sown at the same time and of seeds from plants sown at different times but, harvested at the same time.

Ten plants were selected at random from each collectionsowing combination and seeds were harvested from these same

<sup>-</sup> Harvest times were: (1) 20/8/67; (2) 10/10/67; (3) 24/10/67.

plants at each harvest. The following procedure was adopted to ensure that only mature seeds were collected at each harvest. A polythene bag was placed over the terminal inflorescence of a plant, the inflorescence was gently shaken and any seeds that were dislodged were collected. At the end of the growing season the plants were secured to 6 ft. canes to prevent them from blowing over during the winter (although it was found that this precuation was not necessary as untied plants remained standing). After the winter a final harvest of remaining seeds was made. In this case the entire terminal inflorescence was removed to the laboratory and allowed to dry before the seeds were shaken out.

Seeds collected from individual plants were pooled for each collection-sowing at each harvest. In some collections there was a tendency for the utricles and perianths to persist with the seed. To standardise conditions, seeds of each collection were cleaned by rolling the fruits between two pieces of card with light pressure. The chaff (pieces of utricles, perianth segments and bracts) was separated from the seeds by using a controlled jet of compressed air. While seeds of one treatment were being cleaned, all other seeds were stored in the cold room (see Table 6.3 for conditions).

## 7.3.1.3 Post-harvest treatments

Seven specific post-harvest treatments were used that included four different situations in which the seeds were placed after harvest.

## a) Different wintering situations

Seeds were left in the field in three different situations. In two of these seeds were counted into lots of 200 and placed in small nylon bags (8 x 8 cm) which were sown up with nylon thread. These bags were either buried at a depth of 15 cm below the soil surface or left on the soil surface. In both treatments the bags were secured to small wooden pegs to facilitate their retrieval. Three replicates (bags) of seeds of each treatment were prepared. The use of nylon bags allowed a relatively uninhibited interaction between the seeds and the soil surface. Also the nylon did not decompose during the winter and presumably no chemicals were released that might have influenced the germination behaviour of the seeds.

The third field situation was essentially a pre-harvest treatment as it involved those seeds that persisted on the remains of the parent plant (on-plant) and were harvested after the winter.

In the fourth situation seeds were stored in the <u>laboratory</u> in three replicates of 100 seeds per treatment in glass petri-dishes. The petri-dishes were stored in light-proof aluminum canisters in incubator 13/2 at 10°C. No water was added to the petri-dishes.

# b) Details of specific treatments

Seven specific treatments were included, involving germination trials at three different times. These treatments were as follows:

- 1) Freshly harvested seeds were set to germinate in the greenhouse in October, 1967.
- 2) Bags of seeds of each collection-sowing-harvest were arranged randomly in plots on the soil surface and left there until late November, 1967. Then they were buried until early May, 1968 whereupon the seeds were set to germinate in either the laboratory or the field.
- Bags of seeds were left on the soil surface until May, 1968 whereupon the seeds were set to germinate in either the laboratory or the field.
- 4) Seeds that had persisted on the plant remains in the field were set to germinate in either the laboratory or the field in May, 1968.
- 5) Seeds were stored in petri-dishes in the laboratory until May, 1968 when they were set to germinate in the laboratory.
- Bags of seeds were left on the soil surface until
  November, 1968. They were then buried until May,
  1969 whereupon the seeds were set to germinate in the
  laboratory.
- 7) Bags of seeds were left on the soil surface until May, 1968. They were then buried until May, 1969 whereupon the seeds were set to germinate in the laboratory.

# 7.3.1.4 Treatment combinations

Table 7.3 is a summary of the treatment combinations. In this summary it can be seen that the number of combinations

TABLE 7.3

A SYNOPSIS OF THE COMBINATIONS OF TREATMENTS EXAMINED AT EACH GERMINATION TRIAL IN EXPERIMENT 9

4	Hay, 1969 laboratory	***			****	C x x x x x x x		
on trial	May, 1968 field	***	***		****	****		
Germination trial 2	Mey, 1968 laboratory	~***	***	***	****	****	* * * * *	
~	Oct., 1967 Greenhouse							**
	Collection	まます と な と な と な と	なななな	4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4	サ サ ロ ス ス ス ス ス ス ス ス ス ス ス ス ス ス ス ス ス	F	TAGARE SZAF	면 단 20
	Post-harvest treatment	Buried	Surface	Laboratory	Buried	Surface	Laboratory	None
	Hervest	7			N			2
	Sowing	~					.*	ļ

	****	****			****	××	××				××
***	****	****	* * * * *		****	××	××	××	××		××
				****						* *	
표 교 교 교 교	で で で で で が り い り い い い い い い い い い い い い い い い い	P2 P2 R1 H7	せ 日 日 日 日 日 日 日 日 日 日 日 日 日	Т Ч Ч Ч Н С С С С С С С С С С С С С С С С С С С	다 다 다 다 다 다 다 다 다 다 다 다 다 다 다 다 다 7 7 7 7 7	년 보 건	년 2 7	ዝ대 ፖር	면 면 27	전 전 건 건	F3
)	Buried	Surface	Laboratory	None	On-plant	Buried	Surface	Laboratory	Laboratory	None	On-plant
	N			<b>1</b> 0	₽	~			0	77	4

		***	8 8 8 8 8 8						
××	<b>!</b>	* * *			***	***		***	××
××	<b>:</b> ×	***	***	×××	XXX	***		***	××
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On-plant	Laboratory	Buried	Surface	Laboratory	Buried	Surface	None	On-prant	Buried Surface None
4	~	N			М			<b>4</b>	М

XX

3

1 - Those treatment combinations included are marked with an "x". Notes:

4

<sup>2 - &</sup>quot;xa" indicates that the post-harvest treatment in this case was five months on the soil surface followed by 12 months buried.

of collections, sowings, harvests and post-harvest treatments that were compared was less than the total of all possible combinations. In part this was planned in order to limit the size of the ultimate germination tests. In other cases certain combinations were impossible; these will now be described:

## a) A. hybridus

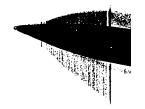
As previously mentioned, only one collection of seeds of this species was available. Moreover, the seeds had been taken from one herbarium sheet and were limited in number. There were sufficient seeds for the first sowing but none for subsequent sowings. It was necessary to raise plants under short days in the laboratory to obtain more seeds and these were not available until the third sowing.

Plants of A. hybridus matured more slowly than those of the other two species, consequently no seeds had been produced from plants of the first sowing in time for the first harvest. In the same way, plants of the third sowing had not produced seeds in time for the third harvest.

Plants of the fourth sowing had produced no mature seeds by the time they were killed by frost.

## b) <u>A. powellii</u>

Because of limitations of space it was decided to use one collection of this species (P3) in all sowings and two further collections (P1 and P2) in the first and third sowings only.



Plants of the third sowing time of P1 were slow to mature and had produced only a few seeds before the third harvest. For the most part, this collection was represented by the first sowing only.

## c) A. retroflexus

At the second harvest no seeds of the third sowing of this species were stored in the laboratory. Unfortunately this procedural error was only discovered at the time the germination trials were conducted.

## 7.3.1.5 Germination trials

## a) Trial 1: October, 1967 - greenhouse

Thirty six small seed boxes (approximately 23 cm by 15 cm by 8 cm deep) were filled to a depth of 5 cm with sterilised potting soil. Three replicates each of 200 seeds of each collection-sowing from harvest three were sown into the soil surface of individual boxes. A small amount of sieved soil was added to cover the seeds to a depth of less than 5 mm. The seed boxes were randomised within a block structure upon a bench in greenhouse 19 (seed Table 6.3) maintained at minimum temperatures of 25°C by day and 15°C by night under a 14-hour daylength. The boxes were watered freely each day of the germination trial.

The experiment was begun on 27th October, 1967 and germination was recorded on alternate days until 20th

November, and then finally on 4th December. A seed was considered to have germinated when the hypocotyl was raised above the soil surface. Once recorded the aerial parts of

a seedling were cut off at the soil surface and removed.

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## b) Trial 2: May, 1968 - laboratory

Seeds retrieved from post-harvest conditions in May,

1968 were cleaned and the replicates of each pooled. From

the seeds of each treatment three replicates each of 50

seeds (100 seeds in the case of laboratory stored seeds)

were counted and placed upon Green's number 450 filter paper

in 90mm glass petri-dishes. Ten millilitres of distilled

water were added to each petri-dish at the beginning of the

trial and further amounts of water were added as needed.

The petri-dishes were arranged on a table in growth room 37D (see Table 6.3) in a randomised block structure. The growth room was maintained under a 14-hour daylength and the initial temperature regime was 25°C by day and 10°C by night. Full details of the environmental conditions in growth room 37D are given in table 6.3.

It was decided to increase the day-time and night-time temperatures as the experiment progressed as a way of observing the effect of higher temperatures on germination. This procedure was adopted since there was not sufficient incubator space available to conduct the experiment simultaneously at different temperatures. The disadvantage of this procedure is that it may not be possible to differentiate between germination as a response to higher temperatures and germination that is naturally delayed at lower temperatures. However the procedure does reflect the general trend between time and temperature that is experienced

in the field during the spring.

After six days of alternating 25°C by day and 10°C by night, the temperature regime was increased by 5C degrees to 30°C and 15°C. After a further six days the day and night temperatures were again increased by 5C degrees and this was repeated after a further six days.

Germination was recorded at daily intervals for the greater part of the trial. A seed was considered to have germinated when its hypocotyl had developed a characteristic red pigmentation. Once a seed had germinated it was removed from the petri-dish. A number of ungerminated seeds remained at the end of 24 days of the experiment. Treatments containing ungerminated seeds were kept under the experimental conditions at 35°C and 20°C, alternating temperatures for a further month after which the water in the petri-dishes was replaced by 10 ml of a 0.1% solution of giberrellic acid. Germination was scored under these conditions for a further two weeks and finally remaining seeds were scarified, using a razor blade to nick the seed coat. One week after scarification a final count was made of germinated seeds. The total germination during the experimental period (24 days) and during the following seven weeks was used as an estimate of viability.

# c) <u>Trial 3: May, 1968 - field</u>

As a comparison with the arbitrary conditions of the laboratory a germination trial was conducted simultaneously under the uncontrolled but more natural conditions of the

Clay flower pots, 15 cm in diameter at the top and field. 13 cm deep were sunk to within 1 cm of their rims in the Guelph loam soil at the Experimental Farm. The pots were filled to within 4 cm of their rims with the soil that had been displaced in sinking them. A one inch layer of the same soil that had been sterilised at 180°F was then added. One hundred and fifty six pots were prepared in this way and arranged in twelve adjacent rows. A known number of seeds of each treatment (usually 150) were scattered on the soil surface in individual pots. Each treatment was replicated three times and the treatments were distributed among the pots in a randomised block arrangement. The soil surface in each pot was then covered with about 5 mm of sterilised soil sieved through a number 7 sieve (Canadian Standard Sieve Series, 2.83 mm opening). After sowing, the soil was thoroughly moistened with a fine spray of water applied from a watering can. The only water that was added subsequently was from natural precipitation.

The experimental design included eighteen pots to which no seeds were added that were intended to monitor the influx of foreign seeds.

The pots were examined at approximately weekly intervals and seedlings were recorded in three categories: those at the cotyledon stage; those with one leaf expanded; and, those with two or more leaves expanded. In this way it was possible to verify that the seedlings counted were of one of the species of Amaranthus. The most similar species

that occurred as a contaminant was Chenopodium rubrum. It did not require much practice to be able to distinguish between Chenopodium rubrum and the three species of Amaranthus at the cotyledon stage.

Observations of germination were continued until the end of July by which time the soil had become strongly caked and little further germination was observed.

No estimate was made of the number of viable seeds remaining at the end of the experiment, and it was feared that seeds had in fact been lost during the experiment through heavy run-off washing through the pots. Ind action may also have removed seeds, although it was noted that there was very little germination of seeds of the three species of Amaranthus in the area between the pots and bounding the experiment. This area was devoid of other vegetation and frequently cultivated by hand to remove weeds.

### d) Trial 4: May, 1969 - laboratory

The germination of seeds that had passed two winters in the field was examined in incubator 13/3 (see Table 6.3) at one temperature regime only, namely 30°C by day and 15°C by night, under a 14-hour day. Germination was scored daily for ten days after which the remaining seeds were scarified with a razor blade to obtain an estimate of viability. In other respects this germination trial was similar to the May, 1968 laboratory trial. In arranging these treatments in the incubator it was not possible to accommodate all of them on the same shelf. This situation complicated the

analysis of results.

#### 7.3.2 Results

The cumulative germination scores for each of the four trials included in this experiment are presented in Appendix 3, tables A3.9 to A3.12

7.3.2.1 Germination trial 1 - October, 1967

#### a) General

The treatments that were included in this germination trial were all from harvest three and although each collection was represented, the number of sowings of each collection differed.

In a preliminary analysis of the results the logical relationship between the same sowing of different collections Seeds from each sowing were considered merely was ignored. as sub-samples of the collection they represented. way the data constituted an unbalanced hierachical classification and could be analysed following the methods outlined The in Appendix 4 for nested or hierachical designs. results of this preliminary analysis are presented in Appendix Germination occurred at a very low rate in 5, page **558**. all treatments and the maximum achieved in any replicate was It was thus considered sufficient to compare germination 8%. totals at completion of the experiment rather than daily totals.

The results of the preliminary analysis indicated a significant variance ratio at the level of collections only. As only one species, A. powellii was represented by more than

one collection it was within this species that collections differed. Further analysis was performed upon the data from the different collections of A. powellii and at this time the logical relationship between sowings of different collections was taken into account. The data constituted a two-factorial design but in order to balance the design it was necessary to omit data of sowings 2 and 4 of collection P3 since the other collections were not represented by these sowings. The details of this analysis are presented in Appendix 5, on page 559.

#### b) Results of statistical analysis

#### i) Differences between sowings

No differences were observed in the percentage of germination between seeds of the same collection from different sowing times.

### ii) Differences between collections

A comparison of the mean percentage germination of seeds of each collection showed that seeds of collection P3 gave significantly higher percentage of germination than seeds of either collections P1 or P2.

7.3.2.2 Germination trial 2 - May, 1968, laboratory

#### a) General

The seeds that were set to germinate at this time were of treatments that formed a completely unbalanced design.

It was thus impossible to make a single analysis of the results. Consequently the data were arranged in several comparisons in order to examine each of the factors included.

Table 7.4 describes these comparisons and indicates the levels of each factor examined in each comparison. Each comparison was analysed statistically.

TABLE 7.4

FACTORS EXAMINED IN EACH OF THE SELECTED COMPARISONS ANALYSED AMONG THE RESULTS OF EXPERIMENT 9, GERMINATION TRIAL 2

Comparison number	Sowing times	Harvest <sup>3</sup> times	Post-harvest trestments	Collections	Statistical design
9/1	(1)	1,2	B,S,L	P1,P2,P3,R1	3-factorial
9/2	(1)	(2)	B,S,P,L	P1,P2,P3,R1, H7	2-factorial
9/3	1,3	(2)	B,S,P,L	P2,P3,R1	3-factorial
9/4	(3)	(3)	B,S,P	P2,P3,R1	2-factorial
9/5	(3)	2,3	B,S	P2,P3,R1	3-factorial
9/6	1,2	(1)	B,S,L	P3,R1	3-factorial
9/7	3 <b>,</b> 4	(3)	B,S	(P3)	2-factorial

#### Motes:

<sup>1 -</sup> Under each factor that was varied, the levels that were included are indicated. Under factors that were held constant, the state of the factor is indicated in parentheses.

<sup>2 -</sup> The post-harvest treatments are: Buried (B), soil surface (S), on-plant (P) and laboratory(L).

<sup>3 -</sup> Seeds from post-harvest treatment "P" (on-plant) were all harvested at time 4 (see page 245).

#### b) Statistical analysis of each comparison

The accumulated totals of germination for each day were analysed for each of the comparisons listed in table 7.5. An analysis of daily totals was preferred to an analysis of the final totals in order to detect differences in the rates of germination and to observe any response to the periodic increases in temperature regime.

The estimates of viability were analysed for those comparisons in which different overwintering treatments were compared. In these comparisons, the effects of overwintering treatments on seeds of the same initial viability could be compared. The value of comparing viabilities in the other comparisons was slight, since the viability was expressed as a percentage of the seeds chosen for the experiment. Seeds were initially chosen if they appeared sound. The proportion of unsound seeds in the total production of seeds by a plant was not determined. Germination totals were expressed as a percentage of the total number of viable seeds in the experiment.

The methods of analysis are described in Appendix 4 and in particular follow the flow chart on page 548. As a matter of standard procedure, the perscentages in each comparison were transformed to the arcsin scale. The analysis of each comparison is described in detail in Appendix 5. The results of the analyses are described in the following pages.

# c) Comparison of viabilities: Differences in viability following different post-harvest treatments

Duncan's multiple range test of the mean viability of seeds in each comparison are presented in table. A5.9 (Appendix 5). The mean values for viability for each treatment are presented in table 7.5.

The only significant differences in viability were between seeds that remained on the plant over winter and all other treatments. Significant differences of this nature were observed in collections P3 and R1. However, the seeds that overwintered on the plant cannot be assumed to have had initially the same viability as the seeds used for other treatments since they were not harvested until after the wintering treatment. The differences that exist between these treatments are summarised on pages 564 to 566.

TABLE 7.5

THE MEAN PERCENTAGE VIABILITY OF SAMPLES OF SEEDS
FROM EACH TREATMENT IN EXPERIMENT 9, TRIAL 2
OnOwing Harvest Collection Buried Surface plant Laboratory

G	77	Calleation	Buried	Surface	plant	Tigooracory
Sowing	Harvest	Collection	Darroa			
1	1	P1 P2 P3 R1	98.7 100.0 96.0 98.7	100.0 97.3 93.3 97.3		95.0 95.7 95.0 97.4 94.3
	2	P1 P2 P3 R1 H7	99.3 99.4 96.0 91.3 96.0	98.0 98.7 93.3 96.7 88.0	e2 0	98.3 100.0 97.3 98.0
	4	P1 P2 P3 R1 H7			82.0 92.7 47.4 98.0 92.7	

				G 0	On-	Tabanatam
Sowing	Harvest	Collection	Buried	Surface	prant	Laboratory
2	1	P3 R1	98.0 92.0	94•7 93•3		96•3 98•7
	2	P3 R1				97•3 99•0
	4	P3 R1			60.7 84.0	
3	1	P3				96.3
•	2	P1 P2 P3 R1	98.0 88.0 98.7	98.0 84.7 100.0		94•3 94•3 92•3
	3	P2 P3 R1	97•5 54•7 97•3	98.7 60.0 96.7		
	4	P2 P3 R1 H7			98.0 42.7 79.3 62.0	
4	3	P3	18.0	8.7		

Note: Values shown are the means for three replicates of the total of germination during the trial and after the trial (following scarification etc.) expressed as a percentage of the total number of seeds used.

# d) Analysis of germination scores: comparisons 9/1 to 9/6

The results of the statistical analyses of these comparisons are presented in Appendix 5, from page **566**. The significance of differences between treatment means, as determined by Duncan's multiple range tests, are presented in table A5.15.

# i) Differences between sowing times

For most treatments, seeds harvested at the same time from plants of different sowings showed no differences in germination response (Figs. 7.1, 7.2A-C, 7.4A&B, 7.5A&B). Of seeds of collection P2 that had been buried, those from

plants of the third sowing gave greater germination at lower temperatures than those from plants of the first sowing (Fig. 7.3A). Seeds from plants of the third sowing of collections P3 and R1 that had wintered on plant remains gave greater germination at 30°/15°C than seeds from plants of the first sowing (Fig. 7.4C, 7.5C). A similar response was seen among laboratory-stored seeds of collection P3 (Fig. 7.4D). This comparison could not be made for seeds of collection R1.

#### ii) <u>Differences between harvests</u>

the first harvest gave greater germination at lower temperatures than seeds collected from the same plants at the second harvest if both sets of seeds had wintered either on or below the soil surface (Fig. 7.6, 7.7). Seeds that had been stored in the laboratory showed no differences in germination response that were related to their time of harvest (Fig. 7.8). No differences in germination behaviour were observed between seeds collected at the second harvest and seeds collected at the third harvest (Figs. 7.9, 7.10).

# iii) Differences between post-harvest treatments

The nature of the post-harvest treatment influenced subsequent germination in all collections (Figs. 7.11, 7.12, 7.13, 7.14). The following generalisations can be made. Seeds that had wintered on plant remains gave much lower germination at lower temperatures than seeds that had been buried or seeds that had wintered on the soil surface. As

the temperatures were increased this difference gradually disappeared in all collections except P1 and P2. With seeds from the first and second harvests, the initial germination was greater among seeds that had wintered on the soil surface than for seeds that had been buried. Seeds that had been stored in the laboratory behaved in a similar way to seeds that had wintered on plant remains in collections P1, P2 and R1.

#### iv) Differences between collections

Many differences were observed between seeds from different collections. These differences are illustrated in figures 7.15 to 7.19.

### e) Analysis of germination scores: comparison 9/7

Seeds from the fourth harvest of the third sowing of collection P3 were mostly inviable (Table 7.5). The average number of viable seeds in the three replicates of one treatment was nine; in another it was three. Such small sample sizes would have rendered statistical tests worthless. Thus, this comparison was not analysed.

# 7.3.2.3 Germination trial 3 - May, 1968, field

The treatments included in the field trial of germination at this time were identical to those included in the laboratory trial with the exception that seeds stored in the laboratory after harvest were not included in the field trial. For the purpose of statistical analysis, the treatments were grouped into logical comparisons as before. With the exception mentioned above, these comparisons were

identical to those made with seeds germinated in the laboratory, and they will be described by the numbers indicated in table 7.5.

### a) Statistical analysis of each comparison

Germination was recorded at nine times throughout a two-month period and analyses were performed on the accumulated totals at each of these times. As final estimates of viability were not available two different parameters were analysed. In the first of these germination was expressed as a percentage of the total number of seeds that had been introduced into each pot and in the second, germination was expressed as a percentage of the final germination total in each pot. Use of the first parameter (which will be referred to as "absolute" germination) assumes (a) equal viability of the seeds of all treatments, and (b) that any loss of seeds from the experimental pots occurred randomly. The second parameter (which provides a measure of the rate of germination) requires no assumptions concerning viability. However, use of this parameter does assume that loss of seeds from pots occurred randomly.

The statistical analyses were performed according to the methods described in Appendix 4. Details of the results of the different stages in the analyses are presented in Appendix 5 beginning on page 567. The final results of the analyses are discussed in the following paragraphs (from Duncan's multiple range tests, table A5.20).

#### i) Differences between sowings

For most treatments seeds harvested at the same time from plants of different sowings showed no differences in absolute germination (Figs. 7.20, 7.21A-C, 7.22A-C). The exceptions were seeds of collection R1 wintering on plant remains and on the soil surface. In this collection seeds from plants of the third sowing gave greater germination than seed from plants of the first sowing at certain times during the trial (Fig. 7.23B&C).

Differences in the rates of germination of seeds from plants of different sowings occurred only among seeds that had wintered on plant remains. In collections P2, P3 and R1 seeds from plants of sowing three germinated earlier in the trial than seeds of sowing one.

### ii) Differences between harvests

Seeds of collection P2, P3 and R1 from different harvests of plants sown at the same time exhibited differences in absolute germination after they had wintered on the soil surface (Fig. 7.25). Seeds from the first harvest gave greater germination than seeds from the second harvest for certain periods during the trial. This difference was seen also among seeds of collection P1 but it was not statistically significant.

Seeds from the first and second harvests of collections P1, P2, P3 and R1 that had been buried exhibited similar differences but there were only significant for collection P1 (Fig. 7.24). No significant differences were observed

between seeds from harvests two and three (Figs. 7.26, 7.27).

Differences in absolute germination were also reflected in differences in the rate of germination. In general, seeds from harvest one germinated earlier than seeds from harvest two.

### iii) Differences between post-harvest treatments

There were several differences in absolute germination between seeds that had received different post-harvest treatments. The details of these differences are illustrated in figures 7.21D, 7.22D, 7.23D and 7.28 to 7.30. In general, seeds that had wintered on plant remains gave the lowest germination. Among seeds from the second harvest, those that had been buried gave greater germination than those that had wintered on the soil surface.

Statistically significant differences in the rate of germination with respect to post-harvest treatments are presented in table A5.20 (Appendix 5). Seeds that had wintered on plant remains were the latest to germinate. Seeds from harvests two and three that had been buried germinated more rapidly than seeds from the same harvests that had wintered on the soil surface.

# iv) Differences between species and collections

Many differences in the absolute germination and the rate of germination were observed between different collections subjected to identical treatments. The details of these differences are presented in figures 7.31 to 7.33 and in table A5.20 (Appendix 5). In most treatments seeds of

collection P2 gave a higher percentage germination than seeds from collections P1, P3 and H7 and also germinated more rapidly than seeds of these collections. Seeds of collection R1 gave greater germination and germinated more rapidly than seeds from collections P1, P3 and H7. 7.3.2.4 Germination trial 4 - May, 1969

#### a) General

harvest treatments were compared in this germination trial.

As previously mentioned not all the treatments of this trial could be accommodated on the same shelf in the incubator.

Table 7.6 describes the treatments included and indicates which treatments were assigned to each of the two incubator shelves. It was again necessary to group treatments into a number of comparisons for analysis; (a) because of the inherently unbalanced design; and, (b) because of possible differences in the environments of the two incubator shelves.

Table 7.7 describes the comparisons examined.

TABLE 7.6

TREATMENT COMBINATIONS INCLUDED IN GERMINATION TRIAL 4 (EXPERIMENT 9)

Harvest	Post-harvest treatment	Collection	Incubato upper	r shelf: lower
1	buried	P1 P2	x x	x
			x	x
. 2	buried	P2 P3	x x x	x
		H7		x (continuéd)
	1		treatment  1 buried P1 P2 P3 R1 2 buried P1 P2 P3 R1	Harvest Post-narvest collection upper treatment upper 1 buried P1 P2 X P3 X R1 X P3 X P

Sowing	Harvest	Post-harvest treatment	Collection	Incubator upoer	shelf: lower
	2	surface/ buried	P1 P2 P3 R1	x x	x
3	2	buried	P2 P3 R1	x x	
		surface/ buried	P2 P3 R1	x x	

#### TABLE 7.7

FACTORS EXAMINED IN EACH OF THE SELECTED COMPARISONS ANALYSED AMONG THE RESULTS OF EXPERIMENT 9, GERMINATION TRIAL 4

Comparison	Sowing times	Harvest times	Post-harvest treatments	Collections	Incubator <sup>3</sup> shelves	Statistical design
9/11 9/12 9/13 9/14 9/15 9/16	(1) 1,3 (1) (1) (1) (1)	1,2 <sup>4</sup> (2) 1,2 1,2 (2) (2)	B,SB <sup>4</sup> B,SB (B) (B) B,SB (B)	(R1) P2,P3,R1 P2,P3,R1 P1,R1 P1,R1 P1,R1	L,U U L L L	2-factorial 3-factorial 2-factorial 2-factorial 2-factorial 1-factorial

#### Notes:

- 1 Under each factor that was varied, the levels that were included are indicated. Under factors that were held constant, the state of the factor is indicated in parentheses.
- 2 The post-harvest treatments are: buried for 17 months (B), and surface for 5 months then buried for 12 months (SB).
- 3 Incubator shelves are: lower (L) and upper (U)
- 4 Although different levels of these factors were included they were not examined in this analysis.

### b) Statistical analysis of each comparison

The accumulated totals of germination for each day were analysed for each of the comparisons listed in table 7.6. Germination was expressed as percentages of the total number of viable seeds in each replicate. The estimates of viability were analysed for those comparisons in which different post-harvest treatments were included. The values for viabilities in other comparisons were not analysed for the reasons given on page 257.

The analyses followed the methods outlined in Appendix

4. As a point of standard procedure, the percentages in
each comparison were transformed to the arcsin scale.

Details of each analysis are presented in Appendix 5, pages

567to 584. Differences between treatments are discussed in
the following paragraphs.

# c) Comparison of viabilities: Differences in viability following different post-harvest treatments

The mean values for the viability of seeds in each treatment are presented in table 7.8. The results of Duncan's multiple range tests of the significance of differences between means are presented in table A5.228.

The only significant difference in viability was among seeds of collection R1 that were set to germinate on the lower shelf of the incubator. Seeds that had passed the first winter on the soil surface had a lower viability than seeds that were buried for the first winter. However, seeds of the same treatments that were set to germinate on the upper shelf of the incubator did not exhibit this difference.

TABLE 7.8

THE MEAN PERCENTAGE VIABILITIES OF SAMPLES OF SEEDS FROM EACH TREATMENT IN EXPERIMENT 9, TRIAL 4

Sowing	Harvest	Collection	Incubator shelf	Buried/ buried	Surface/ buried
1	1	P1 P2 P3 R1	T U T	98.7 100.0 96.0 85.4 80.0	
	2	P1 P2 P3 R1 H7	T U T	100.0 99.2 96.9 90.7 80.0 97.8	97.8 97.8 91.1 70.7 79.6
3	2	P2 P3 R1	U U U	99.6 78.7 88.0	86.2 70.2 84.0

Note: Values shown are means of three replicates, expressed as percentages.

### d) Analysis of germination scores

The results of the statistical analyses are presented in Appendix 5 from page **568**. The significance of differences between treatment means, as determined by Duncan's multiple range tests, are presented in table A5.25 (Appendix 5).

### i) <u>Differences between sowings</u>

No differences in germination behaviour were observed between seeds from plants sown at different times (compare Figs. 7.35A and 7.36A with Figs. 7.35B and 7.36B respectively).

## ii) Differences between harvests

No differences in germination behaviour were observed between seeds collected at different harvests(compare Fig.

7.34 and Fig. 7.35A).

### iii) Differences between post-harvest treatments

No differences in germination behaviour were observed between seeds subjected to different post-harvest treatments (compare Figs. 7.35A and 7.35B with Figs. 7.36A and 7.36B respectively).

### iv) Differences between collections

Differences between the germination behaviour of seeds of different collections are illustrated in figures 7.34 to 7.36. Seeds of collection P2 were the most rapid to germinate regardless of treatment. Seeds of collection P1 gave the lowest total percentages of germination which ranged between 40% and 80%.

## v) Differences between positions in the incubator

There were no significant differences in the germination responses of seeds from any treatment of collection R1 when set to germinate on different shelves of the incubator (Figs. 7.34, 7.35A, 7.36A).

# 7.3.3 Discussion of the results of experiment 9

It was mentioned in section 7.2 that the factors examined in this experiment (and others) were mostly complex interactions of two or more variables. For example, in this experiment seeds that were collected at different harvest times were also subjected to post-harvest treatments of different durations. Thus in order to comment meaningfully upon differences in the percentage of germination of seeds subjected to different treatments, it was first

necessary to determine in what ways the treatments themselves varied. In table 7.9, the components contributing to each treatment have been listed. These will be discussed in the following paragraphs.

TABLE 7.9

COMPONENTS OF VARIATION CONTRIBUTING TO EACH OF THE TREATMENTS INCLUDED IN EXPERIMENT 9

Factor	Levels of the factor included (Treatments)	Gр		npor (Se Gs	90 1	rev	be]	LOW	)		Pt
Collections	P1 P2 P3 R1 H7	a b c d e	a b c d e	а а а ъ с			a b c d e				
Sowings	1 2 3 4	a b c d			a b c d		a b c d				
Harvests	1 2 3			a b c	а Ъ С						a b c
Post-harvest treatments						a a b a a a	a a b a a a	a b c d a e	аарсаа	a a b a a a	a a b a c c

#### Notes:

- 1 Individual components do not vary between treatments of each of the above factors if they are represented by the same letter in each vertical column.
- 2 Key to abbreviations for components:
  - Gp genotypic differences between individual
  - Gc genotypic differences between different
  - Gs genotypic differences between different species

(continued)

- Es Environmental conditions as they impinge on plants sown at different times.
- Eh Environmental conditions as they impinge on seeds maturing at different times.
- M Maturity of the plant at the time of harvest.
- Ph Environmental conditions after harvest as they impinge upon seeds at different positions relative to the soil surface.
- Ps Environmental conditions after harvest as they impinge upon seeds in contact with the soil and upon seeds not in contact with the soil.
- Pr Environmental conditions after harvest as they impinge upon seeds retained on plant remains and upon seeds not retained upon plant remains.
- Pt Environmental conditions after harvest as they impinge upon seeds exposed to post-harvest conditions for different lengths of time.

#### a) The effects of different sowing times

The following variables might have influenced the germination behaviour of seeds collected from plants of different sowings:

- 1) The seeds were collected from different individual plants, probably of different genotypes (Table 7.9 component Gp).
- 2) Environmental conditions were continuously changing throughout the growing season. Plants that began their growth and their flowering at different times passed through the same stages of development under different environmental conditions (Table 7.9 component Es).
- 3) Plants of different ages were at different stages of maturity (they may also have matured differently in

response to environmental differences). Differences in maturity may have been accompanied by physiological differences that influenced the development of the seeds (Table 7.9 component  $\underline{M}$ ).

The germination of freshly harvested seeds (trial 1) revealed no differences between seeds from plants of different sowings. However, the greatest germination observed in any treatment was eight percent. Thus, most seeds appeared to be dormant under the conditions used.

The results of the germination trial in the laboratory in the spring following harvest of the seed confirmed the assumption of initial dormancy. At this time seeds of many treatments germinated at the low alternating temperatures (25° and 10°C) at which freshly harvested seeds did not germinate. Seeds from plants of different ages (first and second sowings) harvested early (first harvest) showed no significant differences in their germination after wintering, regardless of post-harvest treatment or collection (Figs. 7.1, 7.2A). However, among the seeds harvested later in the season (second harvest), those from younger plants (third sowing) germinated more rapidly than those from older plants (first sowing) in several of the combinations of post-harvest treatments and collections that were examined (Figs. 7.3A, 7.40&D, 7.5D). When attention is given to the particular treatments in which this difference was observed, an interesting trend is revealed. It appears that the seeds that exhibited this difference belonged to treatments in

which many seeds were not completely after-ripened.

Moreover, the differences observed appear to reflect

differences in the rate of after-ripening but not differences
in the ultimate response of fully after-ripened seeds.

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For example, in collections P3 and R1 a difference in germination response between seeds from plants of different sowings was observed only among those seeds that had wintered in the laboratory or on plant remains in the field (Figs. 7.40&D, 7.50). The very low percentages of germination at 25/10°C of seeds of these treatments, when compared with seeds that had been buried or had wintered on the soil surface, suggests that seeds of the last-mentioned treatments had progressed further in their after-ripening.

In collection F2, the difference in germination response between seeds of the first and third sowings was observed only among seeds that had been buried (Fig. 7.3A). This is in agreement with the previous observations since seeds of this collection were noticeably slower to after-ripen (e.g. see experiment 3, p.223). Seeds of this collection that had wintered in the laboratory or on plant remains in the field hardly had begun to after-ripen by the time of this germination trial and consequently only slight differences were observed with respect to sowing times (Figs. 7.4C&D).

The trend observed in the germination trial in the laboratory (trial 2) was confirmed in the germination trial in the field (Figs. 7.20, 7.21A-C, 7.22A-C, 7.23A-C). Seeds of collection P2 exhibited similar responses to those

of collections P3 and R1 when set to germinate in the field. It is probable that seeds of collection P2 (especially those that had wintered on plant remains) began to after-ripen rapidly during the initial period in this trial, before conditions were suitable for the germination of any seed.

The importance of the state of after-ripening in a sample of seeds for the expression of differences in germination behaviour with respect to sowing time explains in part why seeds from plants of the first and second sowings did not exhibit differences in germination response. that constituted this comparison were harvested two months earlier than seeds that constituted the comparison between the first and third sowings. Presumably after-ripening would have progressed further among seeds from the earlier hervest (this is discussed in the next section). this explanation is not appropriate for seeds of the first harvest that had wintered in the laboratory or on plant Seeds of these treatments did not remains in the field. appear to have completed their after-ripening since their percentage of germination at 25/10°C was much less than that of seeds from other post-harvest treatments (compare Figs. 7.1A&C with Fig. 7.2A and compare Figs. 7.1B&D with Fig. 7.2B).

A further explanation can be presented to explain the apparent absence of influence by sowing time at the first harvest. The difference in age between plants of the first and second sowings was less than the difference in age between plants of the first and third sowings. Thus if the

maturity of the parent plant was a causative factor, this would have differed more between plants of the first and third sowings (seeds collected at the second harvest) than between plants of the first and second sowings (seeds collected at the first sowing).

Further information is available for seeds of collection At the first harvest seeds of this collection were P3. harvested from plants of sowing three as well as from plants of sowings one and two. Seeds from plants of the three ages were stored in the laboratory and included in the germination trial in the following spring. Seeds from plants of the first and second sowings showed little difference in germination response, but seeds from plants of the third sowing germinated more rapidly than seeds from plants of both the first and second sowings (Fig. 7.2B). This observation lends support to the suggestion that maturity of the plant is the major causative factor in preconditioning, since the same response was observed among seeds harvested over a two-month interval. It is unlikely that environmental conditions were changing in the same direction and at the same rate throughout the entire growing It is more likely, however, that physiological season. changes were progressing in one direction and at a similar rate within plants maturing throughout this period.

### b) Differences related to harvest times

The following variables might have influenced the germination behaviour of seeds collected from the same plants at different times:

- 1) Environmental conditions were continuously changing throughout the growing season. Seeds that matured at different times experienced different environmental conditions during their maturation (Table 7.9, component Eh).
- Plants were at different stages of maturity at successive harvest times. Differences in maturity may have been accompanied by physiological differences that influenced the development of the seed (Table 7.9, component M).
- Seeds that were harvested at different times were set to germinate at the same time. Thus they experienced differences in the duration and conditions of post-harvest treatments (Table 7.9, component Pt).

Seeds from different harvests were not set to germinate immediately following their collection. Differences between seeds of different harvests were first examined in the spring following their collection. At this time (trial 2) seeds of the first harvest germinated more rapidly than seeds of the second harvest following post-harvest treatments on or below the surface of the soil (Figs. 7.6, 7.7). Seeds stored in the laboratory generally did not exhibit this difference, although those of collection R1 did to a small extent (Fig. 7.8). Seeds that had wintered on the plant remains were not included in these comparisons since they were not harvested at different times.

The interaction between harvest time and post-harvest conditions suggests that post-harvest differences may be

largely responsible for the results observed. Seeds from harvest one were exposed to post-harvest conditions for two months longer than seeds from harvest two. Moreover the environmental conditions during the first two months (August and September) were very different from those encountered between October and May, for those seeds that were not stored in the laboratory.

The maturity of the parent plant may also have contributed to ultimate germination response although the behaviour of seeds that wintered in the laboratory suggest this is not the case. Perhaps an effect of plant maturity is indicated in the germination response of seeds of collection R1 that had been stored in the laboratory (although this might also reflect the influence of different durations of dry storage). If the influence of harvest time was an effect of plant maturity, it agreed with effects attributed to maturity that were related to sowing time. Thus seeds from younger plants (either the first harvest or the third sowing) after-ripened more rapidly than seeds from older plants (either the second harvest or the first sowing).

The differences observed in the germination responses in the laboratory (trial 2) were confirmed in the field (trial 3), although the differences were not as pronounced (Figs. 7.24, 7.25) and in some cases not statistically significant (Table A5.20).

No differences were observed in germination behaviour

between seeds of the second and third harvests in either laboratory or field trials (Figs. 7.90&D, 7.10, 7.260&D, 7.27). The small interval in time between these harvests (three weeks) compared with the large interval between the first and second harvests (two months) is probably a sufficient explanation of these results.

Seeds that were maintained in post-harvest treatments for more than a year and set to germinate in the second spring after harvest showed no differences in their response with respect to the time of harvest. The rapidity with which these seeds completed their germination suggested that their after-ripening was complete. However, germination was examined under only one alternating temperature regime (30/15°C) and it is possible that differences might have been encountered in germination at lower temperatures.

### c) The influence of post-harvest treatments

The following variables might have influenced the germination behaviour of seeds that received different post-harvest treatments:

1) Seeds on or below the soil surface had contact with the soil whereas those in the laboratory or on plant remains did not. This difference probably resulted in differences in the temperature and moisture conditions that impinged upon the seeds and in the kinds and numbers of micro-organisms with which they came into contact (Table 7.9, component Ps).

- 2) Seeds that wintered on plant remains and seeds on and below the soil surface may also have experienced differences in their environments that were not so much related to contact with the soil but were related to position relative to the soil surface. For an example, the different effects of air speeds at different heights above the ground in increasing the rate of evaporation of water may have resulted in moisture differences in the environments of seeds on the soil surface and those held above the ground on plant remains. At the same time the environment of seeds below the soil surface may have differed in moisture conditions as a result of its insulation from air currents (Table 7.9, component Ph).
- the post-harvest treatment in which seeds were left on the remains of plants differed from the other post-harvest treatments in several respects. Seeds of this treatment were not harvested at the same time as seeds assigned to other treatments and thus they may not have represented the same types of seeds. The seeds may have differed in any of the ways described for seeds from different harvests (Table 7.9, components Eh, M, and Pt). In addition, seeds that over-wintered on the remains of plants may have been influenced by the presence of the decaying plant remains; seeds assigned to other treatments were not subjected to this influence (Table 7.9, component Pr).

Seeds subjected to different post-harvest treatments were first examined in the laboratory, in the spring following their harvest. An important feature of the germination response was a difference in the rate of germination of seeds that had remained on plants from that of seeds that had wintered on or below the soil surface. collections regardless of sowing time, seeds from plant remains germinated less rapidly than seeds from the other The magnitude of this difference varied two treatments. between the collections and was most pronounced in collection (Figs. 7.11, 7.12, 7.13, 7.14). Seeds that had been stored in the laboratory showed a response similar to seeds that had wintered on plant remains in collections P1, P2 and R1, (Figs. 7.11B&C, 7.13B) and a response similar to seeds that had wintered on or below the soil surface in collections P3 and H7 (Figs. 7.12B&C, 7.14C).

In the germination trial that followed in the field,

(which excluded laboratory-stored seeds) the differences

were maintained between seeds that had wintered on plant

remains and seeds from other post-harvest treatments.

However the magnitude of the differences was less, especially

in collection P2, and this result suggested that the

differences were exclusively related to the rate of after
ripening. Thus seeds placed in experimental conditions in

the field undoubtedly continued to after-ripen in the period

preceding the first set of conditions condusive to germination.

During the first seven days of the germination trial in

the laboratory, seeds from the soil surface germinated more rapidly than seeds from beneath the soil surface in several collections (Figs. 7.11B'D, 7.12A, 7.13A, 7.14B). This difference was most pronounced among seeds from harvest one. In the germination trial conducted in the field a completely different response was noted. Seeds that had been buried germinated more rapidly and achieved greater levels of germination than those from the soil surface.

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These two different responses indicate at least that the nature of the post-harvest treatment may influence the ultimate germination response. However, it is not obvious why the ultimate response should be so different in the two germination trials. An explanation is probably related to differences in the environments provided for the two germination trials and the potential for continued after-ripening of seeds assigned to the germination trial in the field.

At first impression it might seem that the response of seeds that had been stored under laboratory conditions indicated the worthlessness of including such an arbitrary treatment. Certainly the results warn against using artificial treatments to predict behaviour under natural conditions. However, the inclusion of the laboratory—stored seeds in this experiment provided insight into the response of seeds that overwintered on plant remains. Without the inclusion of the laboratory—stored seeds it is possible to suggest that the differences in germination

responses between seeds that had wintered on plant remains and seeds that had wintered on or below the soil surface were the result of differences in harvest times or in the types of seeds collected before and after winter. However, this criticism can be countered since seeds that had been stored in the laboratory were part of the same samples from which seeds that had wintered on or below the soil surface had been prepared, yet their germination responses were statistically no different in many cases from those of seeds that had wintered on plant remains.

### d) <u>Differences between collections</u>

#### i) Within A. powellii

When the germination responses of freshly harvested seeds were examined, the only collection of  $\underline{A}$ . powellii in which any seed germinated was collection P3. After the winter, seeds of all three collections of this species germinated, but differences were observed in their rates of germination in the laboratory. Among seeds that had wintered on or below the soil surface, those of collections P1 and P2 germinated more rapidly than those of collection P3 (Fig. 7.15, 7.16, 7.17). Seeds of collection P1 that had wintered on plant remains germinated more rapidly than those of collection P3 from the same source (Fig. 7.19A). Seeds of both these collections gave much greater percentages of germination than seeds of collection P2 from plant remains (Figs. 7.18D, 7.19A). Among laboratory-stored seeds, those of collection P3 germinated more rapidly than those of P1 and seeds of both of these collections gave much greater

germination than those of collection P2 (Fig. 7.18A-C, Fig. 7.19B).

The relationships between collections of A. powellii in their germination behaviour under incubator conditions were not maintained under field conditions (trial 3). The absolute percentage of germination was greater among seeds of collection P2 than among seeds of collection P3. Seeds of all three collections differed in their rates of germination, germinating in the following order of rapidity: P2, P3, and P1. The differences between these results and those obtained in the laboratory trial are no doubt in part a result of the continuation of after-ripening of the seeds when sown in the field.

The germination responses of collections of A. powellii continued to show differences after two winters in post-harvest conditions; seeds of collection P2 germinated more rapidly than those of collections P1 and P3. Seeds of collection P1 appeared to give incomplete germination under the conditions examined, however these conditions were perhaps slightly different from those experienced by collections P2 and P3 (these latter collections were on the upper shelf in the incubator).

The various results described above demonstrate major differences in the rates of after-ripening of seeds of different collections of <u>A. powellii</u> under various post-harvest treatments. There also appears to be an inherent difference between the rate of germination of fully after-

ripened seeds of collection P2 and that of seeds of the other collections.

# ii) Differences between A. hybridus, A. powellii and A. retroflexus

In each germination trial, variation in the germination behaviour of seeds between collections of A. powellii was equal to or greater than variation between any one collection of A. powellii and the collections of either A. hybridus or A. retroflexus. Furthermore, no clear pattern emerged that would describe the overall relationships between the responses of different species. This situation suggests that the range of conditions examined in these germination trials were within the range permitting germination in each species, and that differences in response resulted from differential rates of after-ripening rather than differences in the requirements of fully after-ripened Seeds of A. hybridus gave very little germination seeds. during the first seven days of the laboratory germination trial (at 25°/10°C) (Figs. 7.16, 7.20). However few treatments included seeds of this species and thus this difference could not be confirmed. This species was also the slowest to germinate in the field trial.

Another trend that was observed was that seeds of collections of A. powellii tended to germinate more rapidly than those of Amaranthus retroflexus. However this trend varied considerably depending upon the collection of A. powellii considered, and upon the treatments to which the seeds were subjected.

# e) Comparison of the results of this experiment with the results of previous experiments

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In this experiment and in experiment 1 the germination responses of seeds of collections P1 and R1 can be compared. Seeds of collection P1 were freshly harvested when examined in experiment 1 and very few of them germinated. Freshly harvested seeds of collection P1 gave a very low percentage of germination in trial 1 of this experiment, which was conducted under the same temperature conditions as experiment 1. Thus these two experiments confirm that seeds of this collection are nearly all dormant if they are put to germinate at alternating temperatures of 25° and 10°C, when freshly harvested.

Seeds of collection R1 gave a greater percentage of germination in experiment 1 than in trial 1 of this experiment. This difference may be related to the conditions under which the two experiments were conducted; in experiment 1 seeds were placed in petri-dishes arranged in an incubator and in trial 1 of this experiment seeds were sown on potting soil in small boxes in the greenhouse. It is perhaps significant that there was greater germination among seeds of collection R1 than among seeds of collection P1 in both of these experiments. This suggests that collection P1 has more seeds that are dormant under both of these conditions than collection R1.

Germination of seeds in experiment 3 can be compared with the germination of laboratory-stored seeds in trial 2 of this experiment. Although the temperature regimes

differed between these two experiments, the germination responses showed similarities. The most noticeable similarity was the very low percentage of germination among seeds of collection P2. In addition, the germination responses of collections P1, P3 and R1 were similar both in experiment 3 and in trial 2 of this experiment. In general, the rate of germination of seeds of each collection was higher in experiment 3 than in this experiment. This probably reflects the differences in temperatures between the two experiments.

The germination of seeds of collection P5 in experiment 7 can be compared with the germination of seeds of collection P2 from laboratory-storage and plant remains in trial 2 of this experiment. The justification for comparing collections P2 and P5 is based upon the similarity of response between seeds of these two collections in The germination response of experiments 3 and 10. laboratory-stored seeds was similar in both experiments; more than 75% of seeds were dormant under the conditions examined. Seeds that had over-wintered on plant remains gave a much higher germination in experiment 7 than in this The seeds used in experiment 7 were collected experiment. three weeks later in the season than those used in this experiment and the two collections were made in different The differences probably reflect different degrees years. of after-ripening in the two collections of seed.

#### TABLE 7.10

#### A KEY TO THE SYMBOLS USED IN FIGURES 7.1 TO 7.19

#### 1) Temperature regimes

The temperature regime at which the experiment was conducted was raised periodically. The sequence of temperatures is indicated in each figure by a horizontal band of different patterns:

PATTERN		<b>******</b>		
TEMPERATURE: by day	25	30	35	40
by night (°C)	10	15	20	25

#### 2) Species and collections

Different species are represented by different symbols that serve as points on the graphs. In figures that compare the germination of seeds of different species or collections from identical treatments the lines joining points are different for different species. Collections are identified by numbers within the symbols. In figures that compare the germination of seeds of the same collection from different treatments the collection is indicated by a small symbol at the right side of the graph.

Species	${ t Symbols}$	Cottecrious
A. hybridus	▼	H7
A. powellii	Δ	P1, P2, P3
A. retroflexus	0	Rl

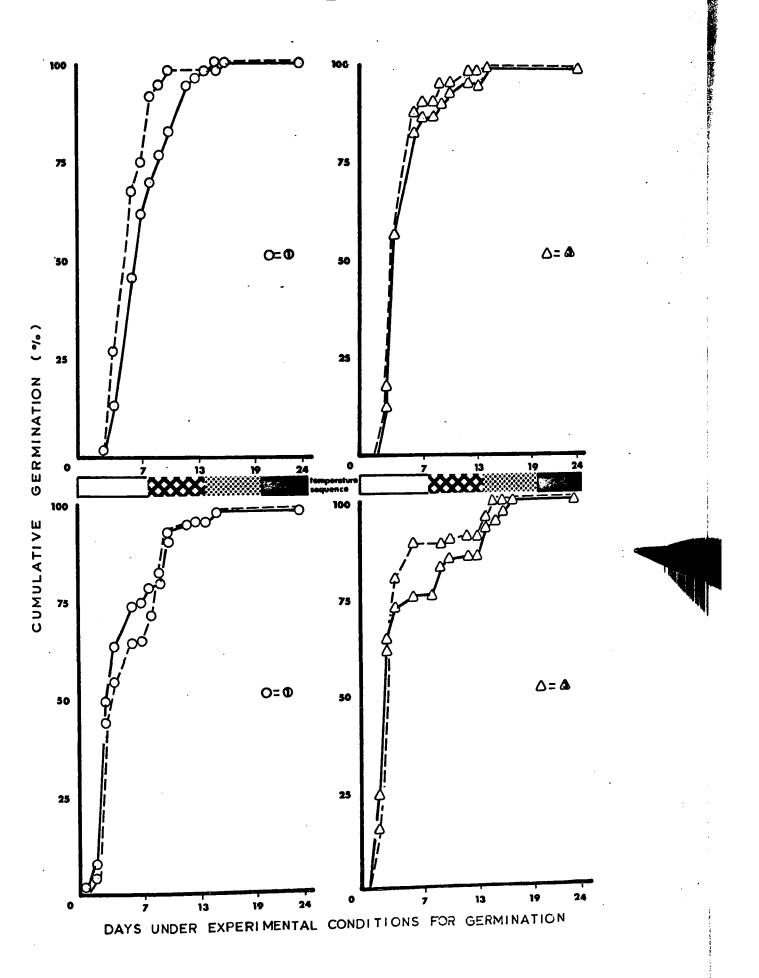
#### 3) Treatments

In figures that compare the germination of seeds from different treatments lines joining points on the graphs are different and indicate the different treatmenents:

(continued)

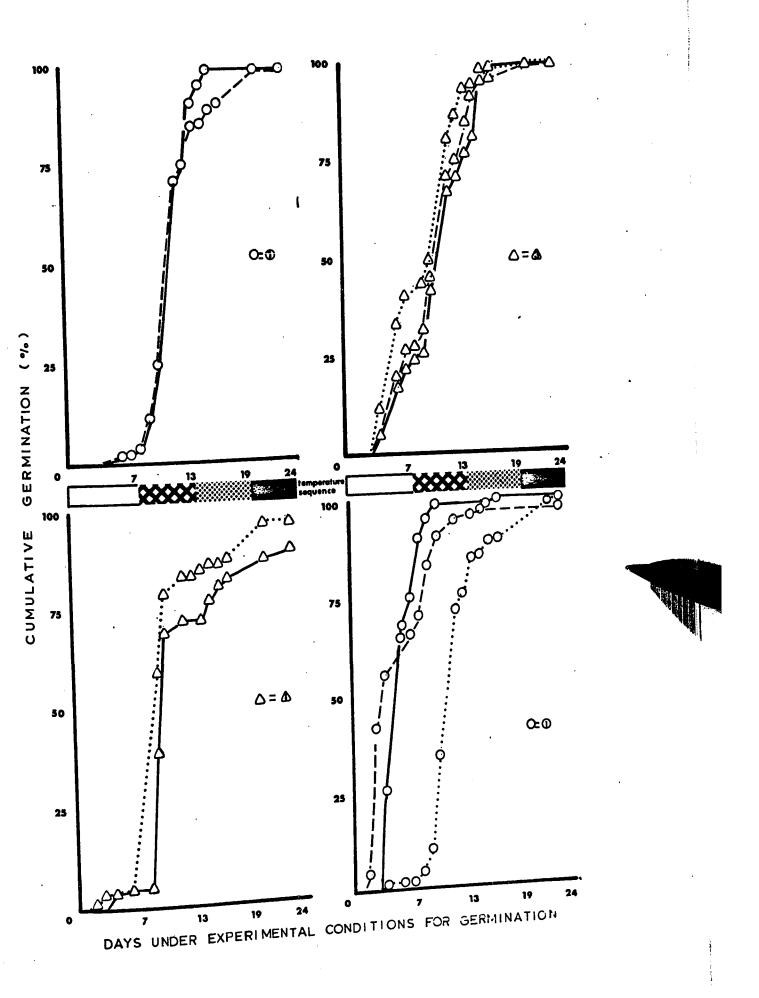
sowing:	1	2	3	
SYMBOL:	-		•••••	
HARVEST:	1	2	3	
SYMBOL:	<del></del>	***************************************	•••••	
POST-HARVEST TREATMENT:	BURIED	SURFACE	ON-PLANT LABORATORY	
SYMBOL:				

- Figure 7.1A A comparison of the germination, after wintering beneath the soil, of seeds of collection R1 collected at harvest one from plants of two different sowings.
- Figure 7.1B A comparison of the germination, after wintering beneath the soil, of seeds of collection P3 collected at harvest one from plants of two different sowings.
- Figure 7.10 A comparison of the germination, after wintering on the soil surface, of seeds of collection R1 collected at harvest one from plants of two different sowings.
- Figure 7.1D A comparison of the germination, after wintering on the soil surface, of seeds of collection P3 collected at harvest one from plants of two different sowings.





- Figure 7.2A A comparison of the germination, after wintering in the laboratory, of seeds of collection R1 collected at harvest one from plants of two different sowings.
- Figure 7.2B A comparions of the germination, after wintering in the laboratory, of seeds of collection P3 collected at harvest one from plants of three different sowings.
- Figure 7.2C A comparison of the germination, after wintering in the laboratory, of seeds of collection P1 collected at harvest one from plants of two different sowings.
- Figure 7.2D A comparison of the germination, after wintering in three different situations, of seeds of collection R1 collected at harvest one from plants of sowing two.



- Figure 7.3A A comparison of the germination, after wintering beneath the soil, of seeds of collection P2 collected at harvest two from plants of two different sowings.
- Figure 7.3B A comparison of the germination, after wintering on the soil surface, of seeds of collection P2 collected at harvest two from plants of two different sowings.
- Figure 7.30 A comparison of the germination, after wintering on plant remains, of seeds of collection P2 collected from plants of two different sowings.
- Figure 7.3D A comparison of the germination, after wintering in the laboratory, of seeds of collection P2 collected at harvest two from plants of two different sowings.

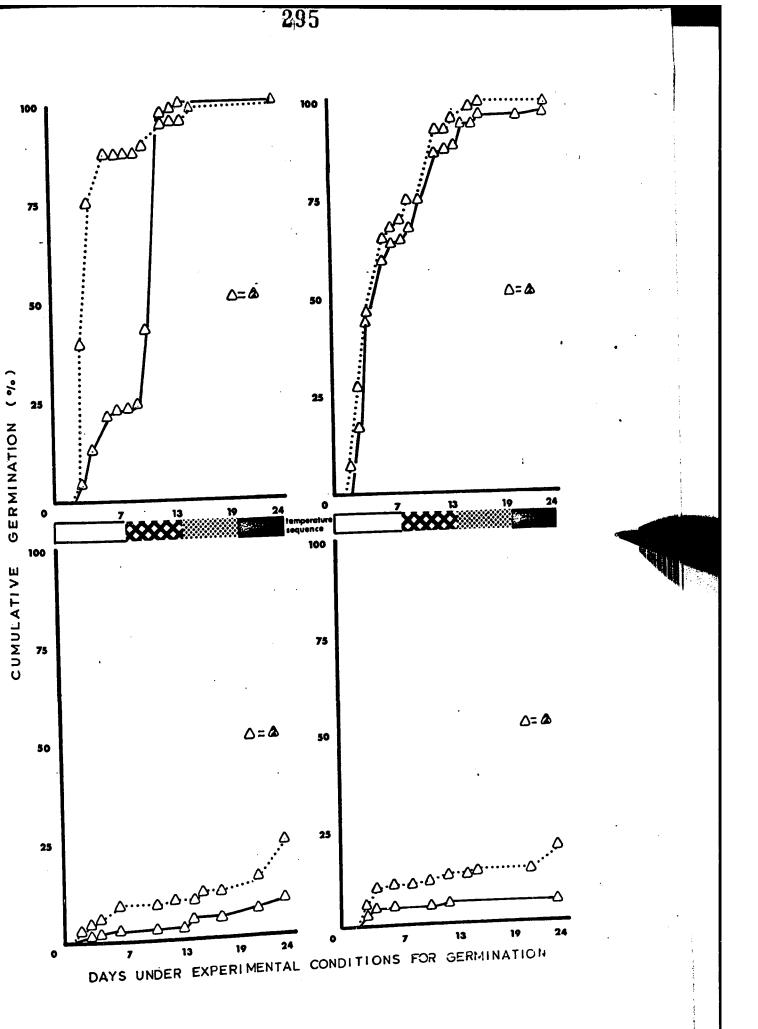




Figure 7.4A A comparison of the germination, after wintering beneath the soil, of seeds of collection P3 collected at harvest two from plants of two different sowings.

Figure 7.4B A comparison of the germination, after wintering on the soil surface, of seeds of collection P3 collected at harvest two from plants of two different sowings.

Figure 7.4C A comparison of the germination, after wintering on plant remains, of seeds of collection P3 collected from plants of two different sowings.

Figure 7.4D A comparison of the germination, after wintering in the laboratory, of seeds of collection P3 collected at harvest two from plants of two different sowings.

Figure 7.5A A comparison of the germination, after wintering beneath the soil, of seeds of collection R1 collected at harvest two from plants of two different sowings.

Figure 7.5B A comparison of the germination, after wintering on the soil surface, of seeds of collection R1 collected at harvest two from plants of two different sowings.

Figure 7.50 A comparison of the germination, after wintering on plant remains, of seeds of collection R1 collected from plants of two different sowings.

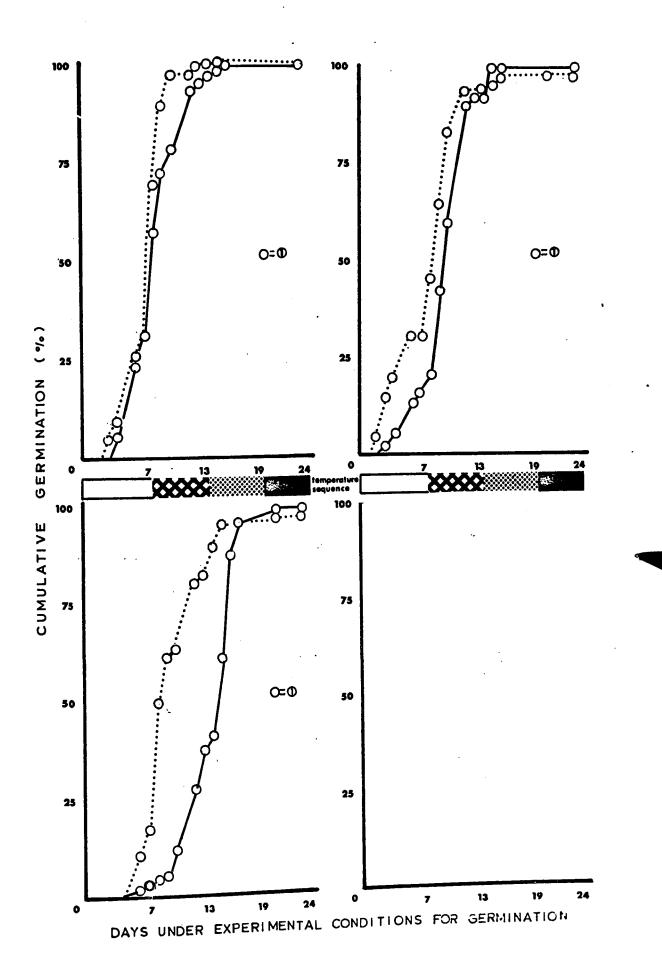




Figure 7.6A A comparison of the germination, after wintering beneath the soil, of seeds of collection R1 collected at two different harvests from plants of sowing one.

Figure 7.6B A comparison of the germination, after wintering beneath the soil, of seeds of collection P1 collected at two different harvests from plants of sowing one.

Figure 7.60 A comparison of the germination, after wintering beneath the soil, of seeds of collection P2 collected at two different harvests from plants of sowing one.

Figure 7.6D A comparison of the germination, after wintering beneath the soil, of seeds of collection P3 collection at two different harvests from plants of sowing one.

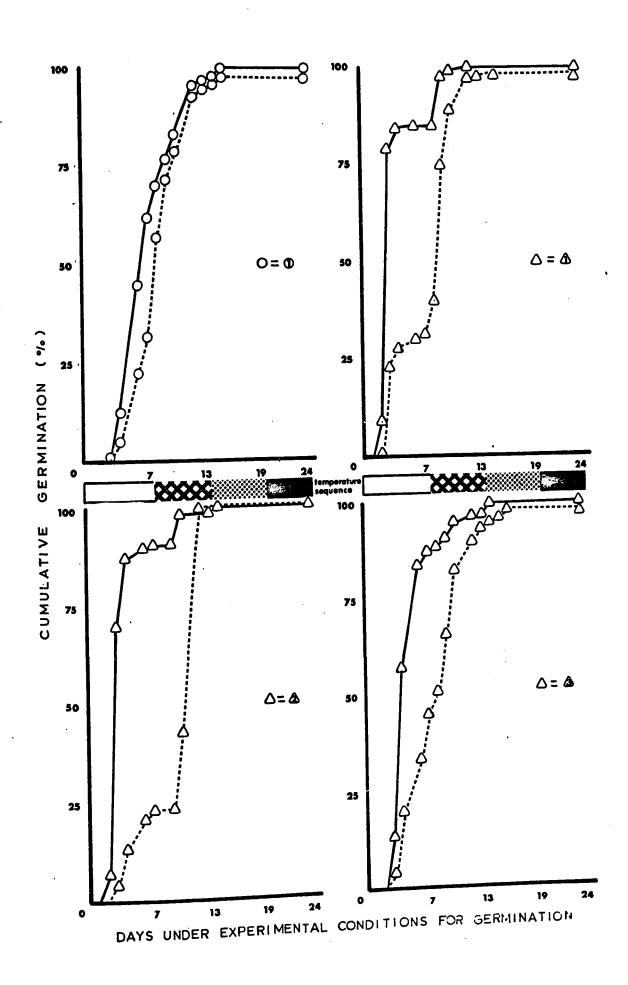
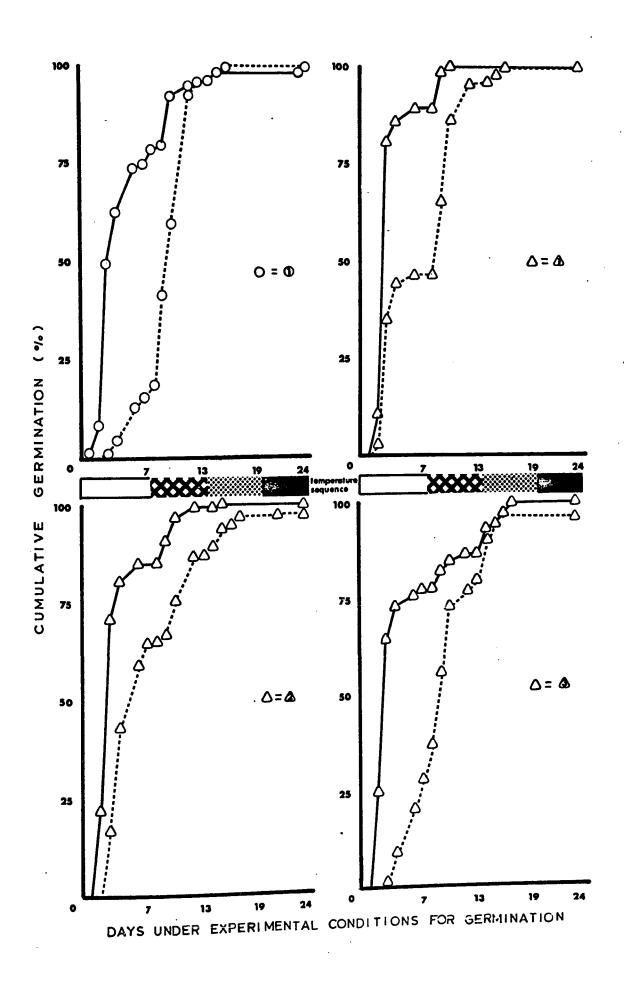


Figure 7.7A A comparison of the germination, after wintering on the soil surface, of seeds of collection R1 collected at two different harvests from plants of sowing one.

Figure 7.7B A comparison of the germination, after wintering on the soil surface, of seeds of collection P1 collected at two different harvests from plants of sowing one.

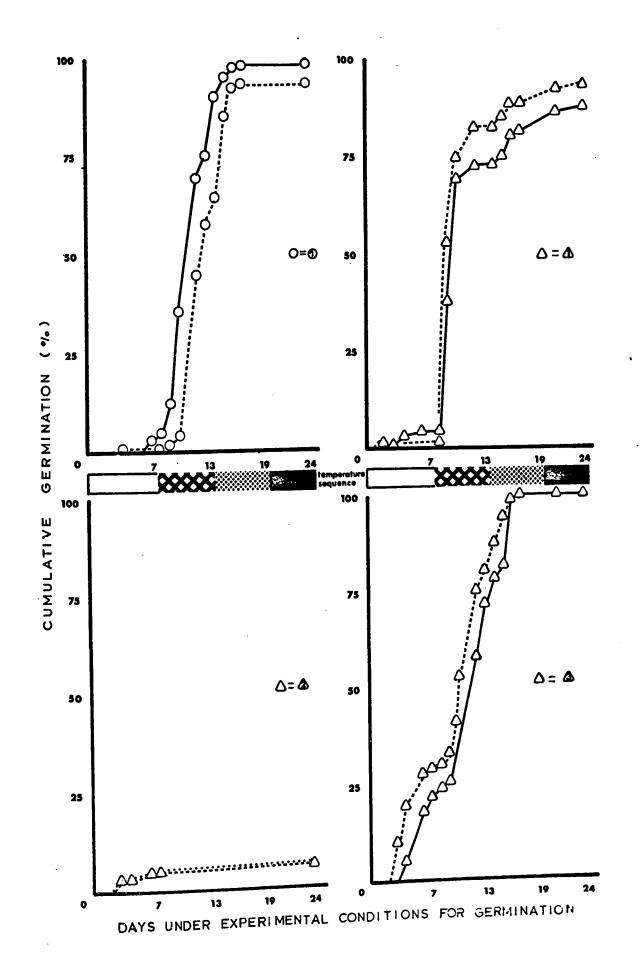
Figure 7.70 A comparison of the germination, after wintering on the soil surface, of seeds of collection P2 collected at two different harvests from plants of sowing one.

Figure 7.7D A comparison of the germination, after wintering on the soil surface, of seeds of collection P3 collected at two different harvests from plants of sowing one.

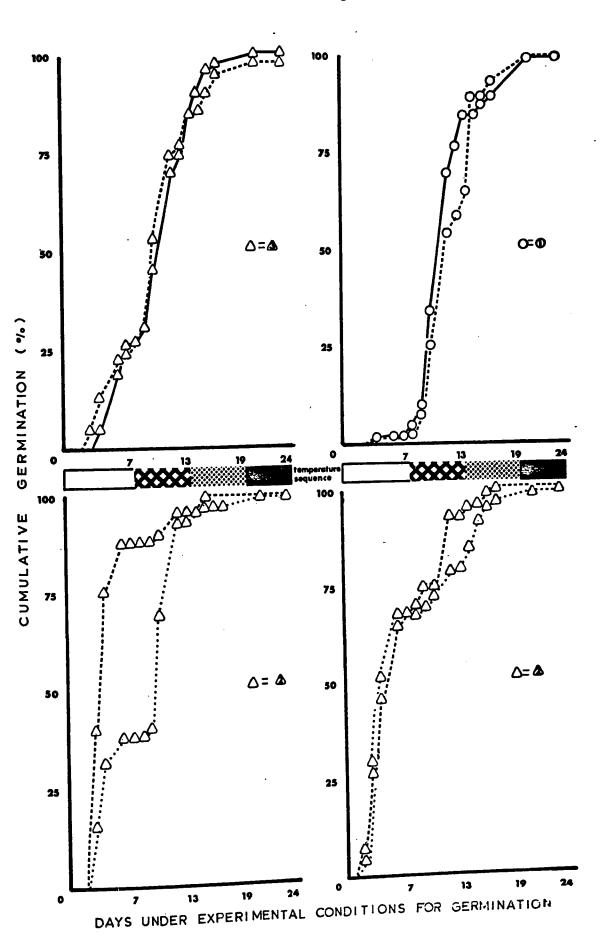




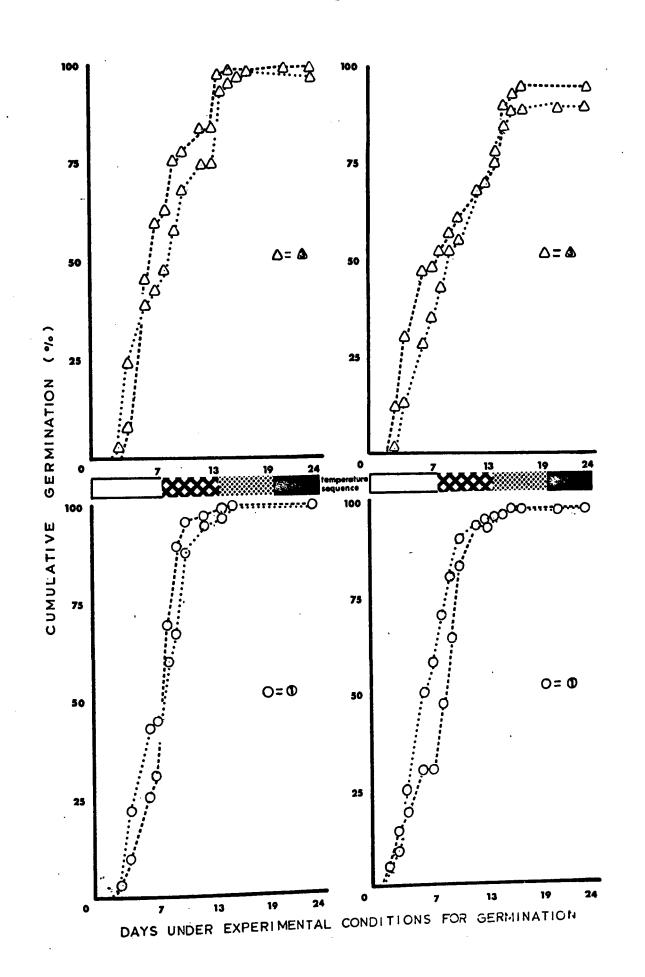
- Figure 7.8A A comparison of the germination, after wintering in the laboratory, of seeds of collection R1 collected at two different harvests from plants of sowing one.
- Figure 7.8B A comparison of the germination, after wintering in the laboratory, of seeds of collection P1 collected at two different harvests from plants of sowing one.
- Figure 7.8C A comparison of the germination, after wintering in the laboratory, of seeds of collection P2 collected at two different harvests from plants of sowing one.
- Figure 7.8D A comparison of the germination, after wintering in the laboratory, of seeds of collection P3 collected at two different harvests from plants of sowing one.



- Figure 7.9A A comparison of the germination, after wintering in the laboratory, of seeds of collection P3 collected at two different harvests from plants of sowing two.
- Figure 7.9B A comparison of the germination, after wintering in the laboratory, of seeds of collection R1 collected at two different harvests from plants of sowing two.
- Figure 7.90 A comparison of the germination, after wintering beneath the soil, of seeds of collection P2 collected at two different harvests from plants of sowing three.
- Figure 7.9D A comparison of the germination, after wintering on the soil surface, of seeds of collection P2 collected at two different harvests from plants of sowing three.



- Figure 7.10A A comparison of the germination, after wintering beneath the soil, of seeds of collection P3 collected at two different harvests from plants of sowing three.
- Figure 7.10B A comparison of the germination, after wintering on the soil surface, of seeds of collection P3 collected at two different harvests from plants of sowing three.
- Figure 7.10C A comparison of the germination, after wintering beneath the soil, of seeds of collection R1 collected at two different harvests from plants of sowing three.
- Figure 7.10D A comparison of the germination, after wintering on the soil surface, of seeds of collection R1 collected at two different harvests from plants of sowing three.





- Figure 7.11A A comparison of the germination, after wintering in three different situations, of seeds of collection P2 collected at harvest one from plants of sowing one.
- Figure 7.11B A comparison of the germination, after wintering in <u>four different situations</u>, of seeds of collection P2 collected at harvest 2\* from plants of sowing one.
- Figure 7.11C A comparison of the germination, after wintering in <u>four different situations</u>, of seeds of collection P2 collected at harvest two\* from plants of sowing three.
- A comparison of the germination, after wintering in three different situations, of seeds of collection P2 collected at harvest three\* from plants of sowing three.
- \*Mote: Seeds that overwintered on plant remains were harvested after winter at harvest four.

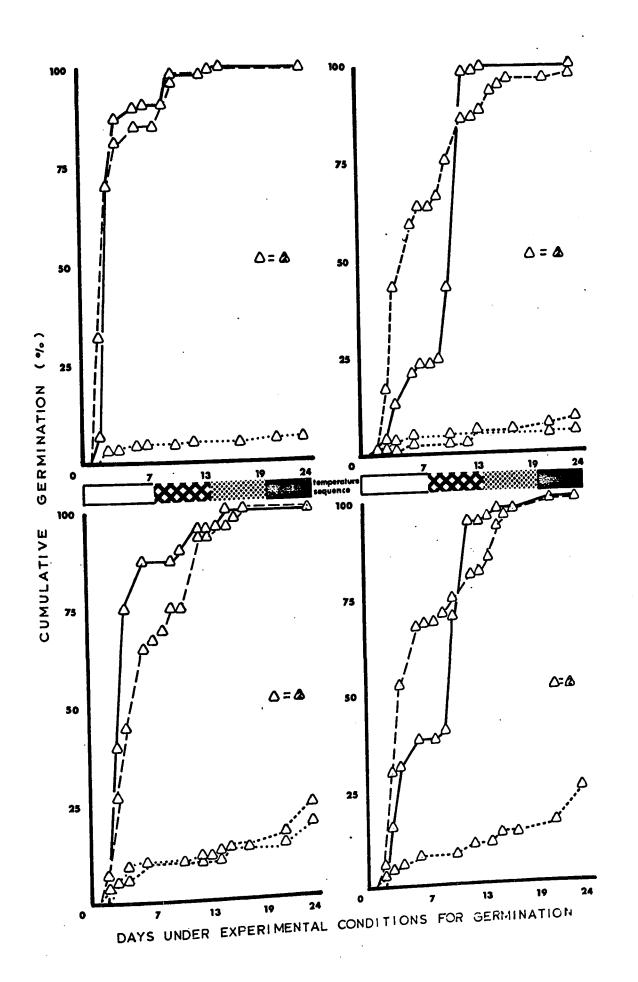


Figure 7.12A A comparison of the germination, after wintering in three different situations, of seeds of collection P3 collected at harvest one from plants of sowing one.

Figure 7.12B A comparison of the germination, after wintering in four different situations, of seeds of collection P3 collected at harvest two\* from plants of sowing one.

Figure 7.12C A comparison of the germination, after wintering in four different situations, of seeds of collection P3 collected at harvest two\* from plants of sowing three.

Figure 7.12D A comparison of the germination, after wintering in three different situations, of seeds of collection P3 collected at harvest three\* from plants of sowing three.

\*Note: Seeds that overwintered on plant remains were harvested after winter at harvest four.

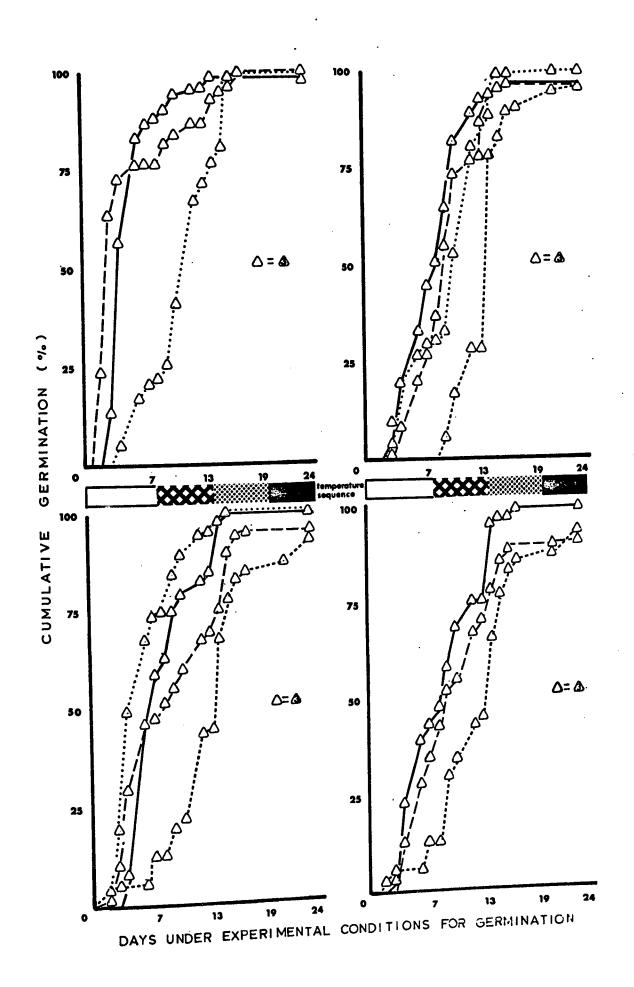


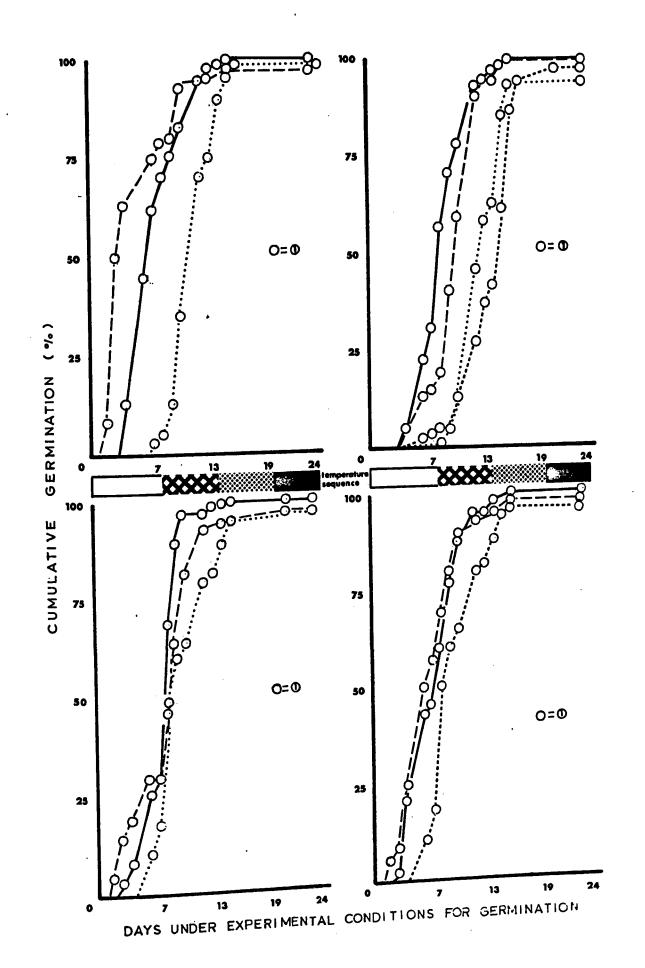
Figure 7.13A A comparison of the germination, after wintering in three different situations, of seeds of collection R1 collected at harvest one from plants of sowing one.

Figure 7.13B A comparison of the germination, after wintering in four different situations, of seeds of collection R1 collected at harvest two\* from plants of sowing one.

Figure 7.130 A comparison of the germination, after wintering in <u>four different situations</u>, of seeds of collection R1 collected at harvest two\* from plants of sowing three.

Figure 7.13D A comparison of the germination, after wintering in three different situations, of seeds of collection R1 collected at harvest three\* from plants of sowing three.

\*Note: Seeds that overwintered on plant remains were harvested after winter at harvest four.





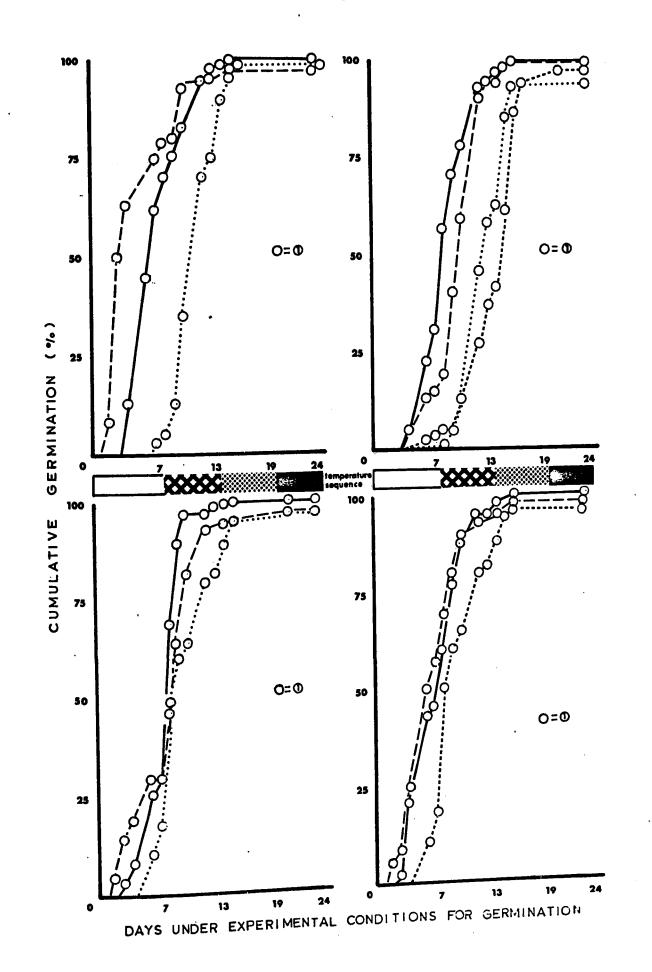




Figure 7.14A A comparison of the germination, after wintering in three different situations, of seeds of collection P1 collected at harvest one from plants of sowing one.

Figure 7.14B A comparison of the germination, after wintering in <u>four different situations</u>, of seeds of collection P1 collected at harvest two\* from plants of sowing one.

Figure 7.14C A comparison of the germination, after wintering in four different situations, of seeds of collection H7 collected at harvest two\* from plants of sowing one.

Figure 7.14D A comparison of the germination, after wintering in three different situations, of seeds of collection P3 collected at harvest one from plants of sowing two.

\*Note: Seeds that overwintered on plant remains were harvested after winter at harvest four.

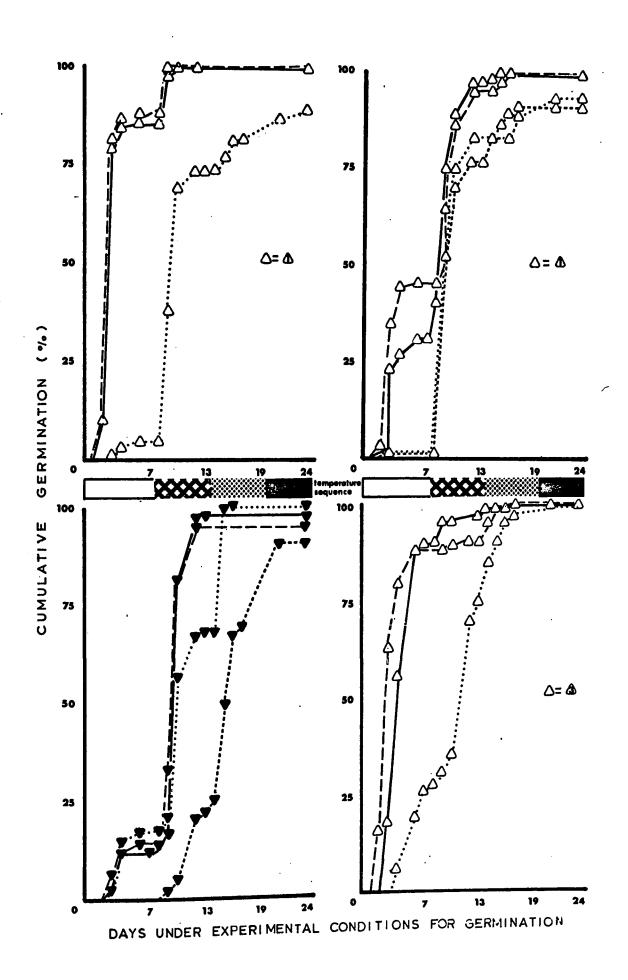




Figure 7.15A A comparison of the germination, after wintering beneath the soil, of seeds of four different collections collected at harvest one from plants of sowing one.

Figure 7.15B A comparison of the germination, after wintering beneath the soil, of seeds of two different collections collected at harvest one from plants of sowing two.

Figure 7.150 A comparison of the germination, after wintering beneath the soil, of seeds of three different collections collected at harvest two from plants of sowing three.

Figure 7.15D A comparison of the germination, after wintering beneath the soil, of the seeds of three different collections collected at harvest three from plants of sowing three.

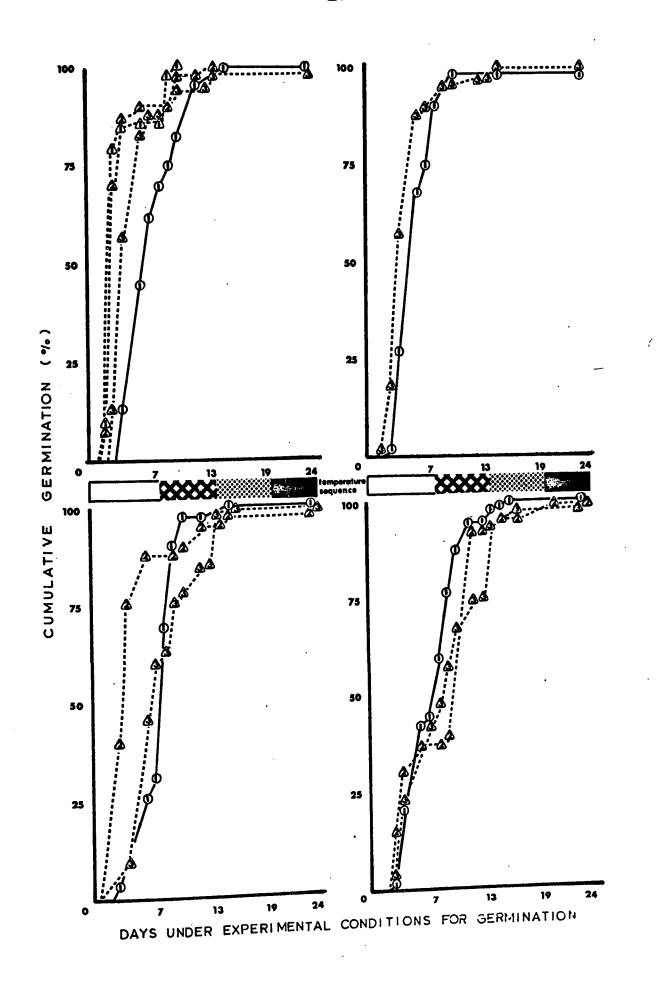




Figure 7.16A A comparison of the germination, after wintering beneath the soil, of seeds of five different collections collected at harvest two from plants of sowing one.

Figure 7.16B A comparison of the germination, after wintering on the soil surface, of seeds of five different collections collected at harvest two from plants of sowing one.

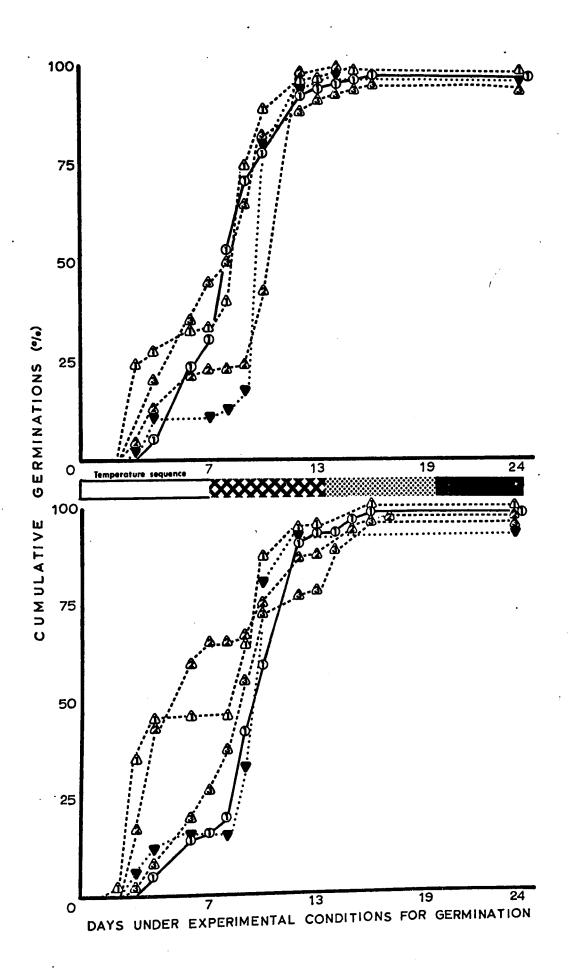




Figure 7.17A A comparison of the germination, after wintering on the soil surface, of seeds of four different collections collected at harvest one from plants of sowing one.

Figure 7.17B A comparison of the germination, after wintering on the soil surface, of seeds of two different collections collected at harvest one from plants of sowing two.

Figure 7.17C A comparison of the germination, after wintering on the soil surface, of seeds of three different collections collected at harvest two from plants of sowing three.

Figure 7.17D A comparison of the germination, after wintering on the soil surface, of the seeds of three different collections collected at harvest three from plants of sowing three.

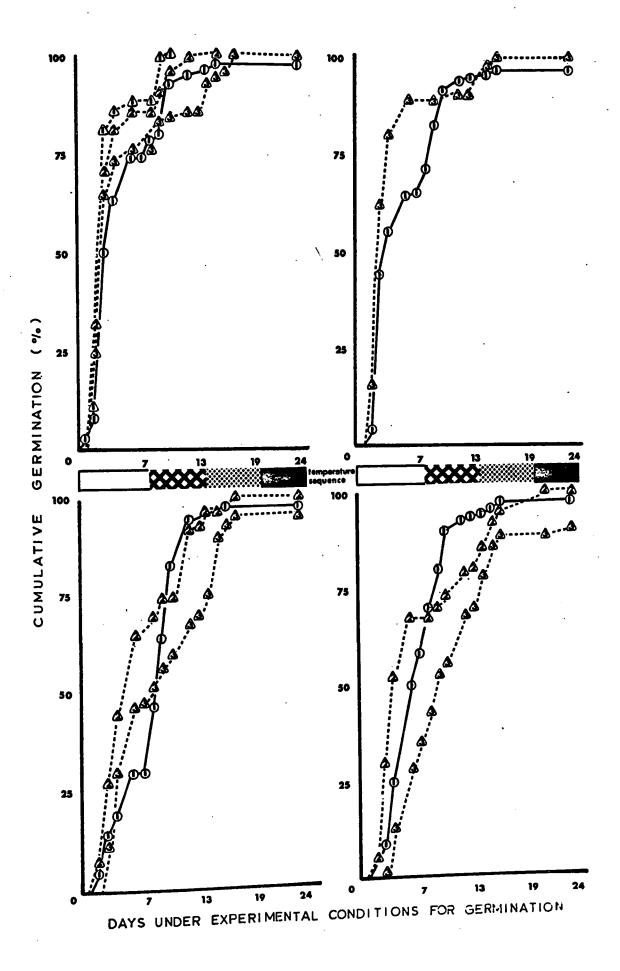




Figure 7.18A A comparison of the germination, after wintering in the laboratory, of seeds of four different collections collected at harvest one from plants of sowing one.

Figure 7.18B A comparison of the germination, after wintering in the laboratory, of seeds of two different collections collected at harvest one from plants of sowing two.

Figure 7.18C A comparison of the germination, after wintering in the laboratory, of seeds of two different collections collected at harvest two from plants of sowing three.

Figure 7.18D A comparison of the germination, after wintering on plant remains, of seeds of three different collections from plants of sowing three.

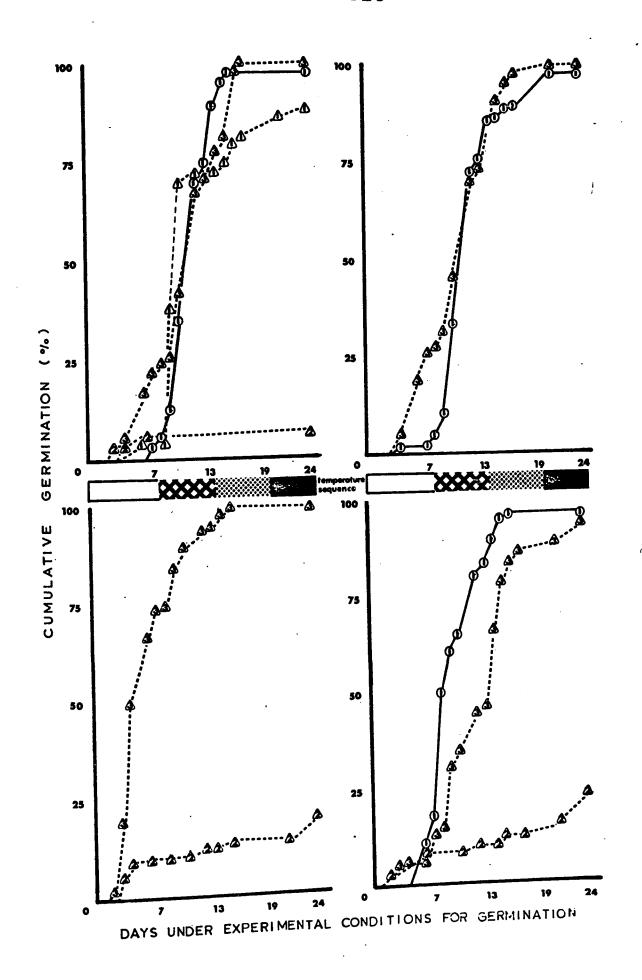




Figure 7.19A A comparison of the germination, after wintering on plant remains in the field, of seeds of <u>five different collections</u> from plants of sowing one.

Figure 7.19B A comparison of the germination, after wintering in the laboratory, of seeds of five different collections collected at harvest two from plants of sowing one.

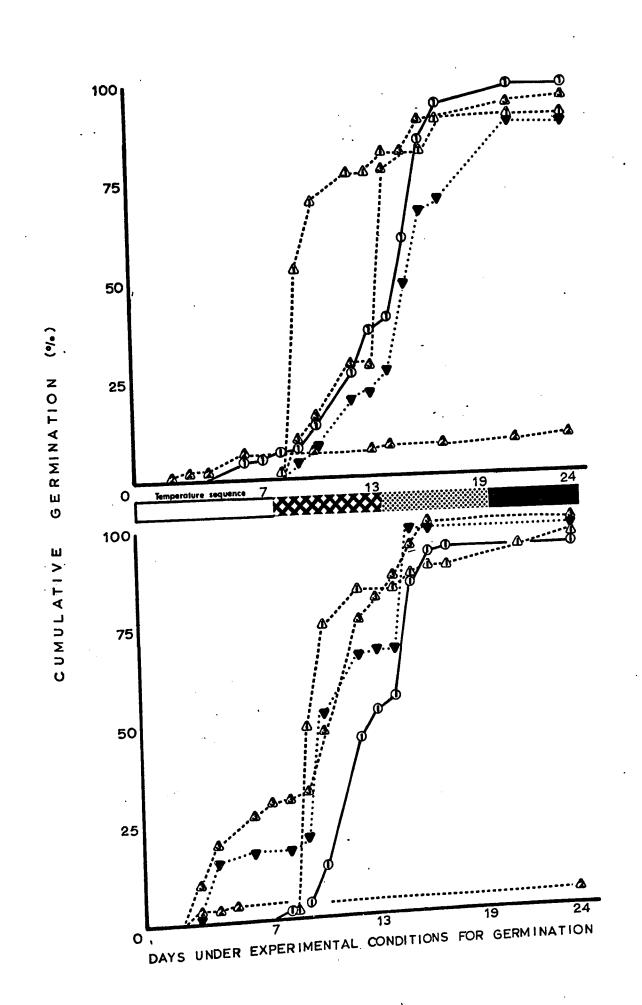




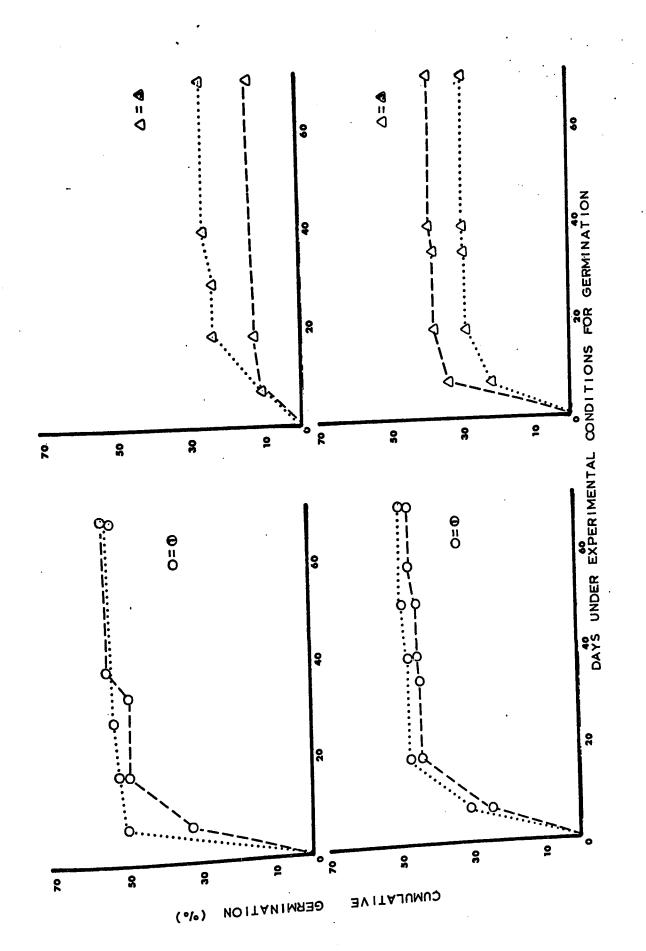
Figure 7.20A A comparison of the germination (in the field), after wintering beneath the soil, of seeds of collection R1 collected at harvest one from plants of two different sowings.

Figure 7.20B A comparison of the germination (in the field), after wintering beneath the soil, of seeds of collection P3 collected at harvest one from plants of two different sowings.

Figure 7.20C A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection R1 collected at harvest one from plants of two different sowings.

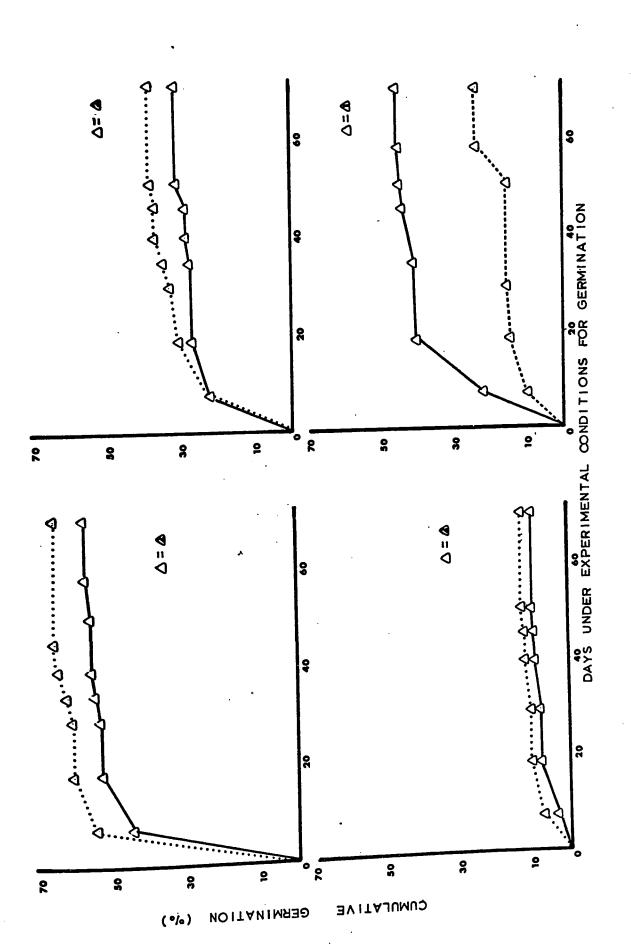
Figure 7.20D A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection P3 collected at harvest one from plants of two different sowings.

Notes: 1 - In each of these figures germination is expressed as a percentage of the total number of seeds nown



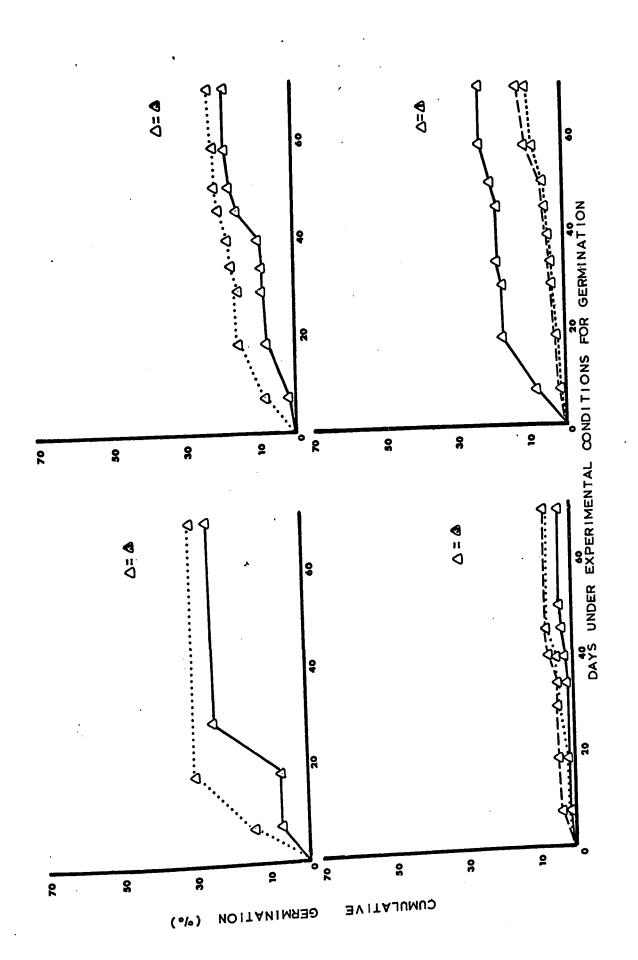


- Figure 7.21A A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of collection P2 collected at harvest two from plants of two different sowings.
- Figure 7.21B A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection P2 collected at harvest two from plants of two different sowings.
- Figure 7.21C A comparison of the germination (in the field), after wintering on plant remains, of seeds of collection P2 from plants of two different sowings.
- Figure 7.21D A comparison of the germination (in the field), after wintering in two different situations, of seeds of collection P1 collected at harvest one from plants of sowing one.
- Notes: 1 In each of these figures germination is expressed as a percentage of the toal number of seeds sown.
  - 2 In order to interpret these figures consult table 7.10 for a key to the symbols used.



- Figure 7.22A A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of collection P3 collected at harvest two from plants of two different sowings.
- Figure 7.22B A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection P3 collected at harvest two from plants of two different sowings.
- Figure 7.22C A comparison of the germination (in the field), after wintering on plant remains, of seeds of collection P3 from plants of two different sowings.
- Figure 7.22D A comparison of the germination (in the field), after wintering in three different situations, of seeds of collection P1 collected at harvest two\* from plants of sowing one.
- \*Notes: 1 Seeds that overwintered on plant remains were harvested after winter at harvest four.
  - 2 In each of these figures germination is expressed as a percentage of the total number of seeds sound
  - 3 In order to interpret these figures consult table 7.10 for a key to the symbols used.

- Figure 7.22A A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of collection P3 collected at harvest two from plants of two different sowings.
- Figure 7.22B A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection P3 collected at harvest two from plants of two different sowings.
- Figure 7.220 A comparison of the germination (in the field), after wintering on plant remains, of seeds of collection P3 from plants of two different sowings.
- Figure 7.22D A comparison of the germination (in the field), after wintering in three different situations, of seeds of collection P1 collected at harvest two\* from plants of sowing one.
- \*Notes: 1 Seeds that overwintered on plant remains were harvested after winter at harvest four.
  - 2 In each of these figures germination is expressed as a percentage of the total number of seeds soun.
  - 3 In order to interpret these figures consult table 7.10 for a key to the symbols used.



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- Figure 7.23A A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of collection R1 collected at harvest two from plants of two different sowings.
- Figure 7.23B A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection R1 collected at harvest two from plants of two different sowings.
- Figure 7.23C A comparison of the germination (in the field), after wintering on plant remains, of seeds of collection R1 from plants of two different sowings.
- Figure 7.23D A comparison of the germination (in the field), after wintering in three different situations, of seeds of collection H7 collected at harvest two\* from plants of sowing one.
- \*Notes: 1 Seeds that overwintered on plant remains were harvested after winter at harvest four.
  - 2 In each of these figures germination is expressed as a percentage of the total number of seeds sown.
  - 3 In order to interpret these figures consult table 7.10 for a key to the symbols used.

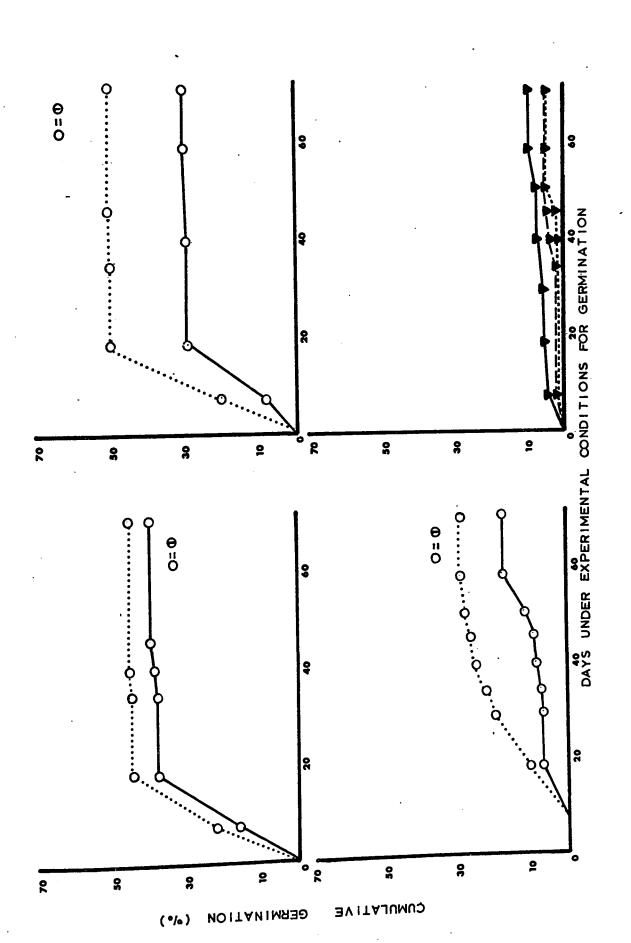




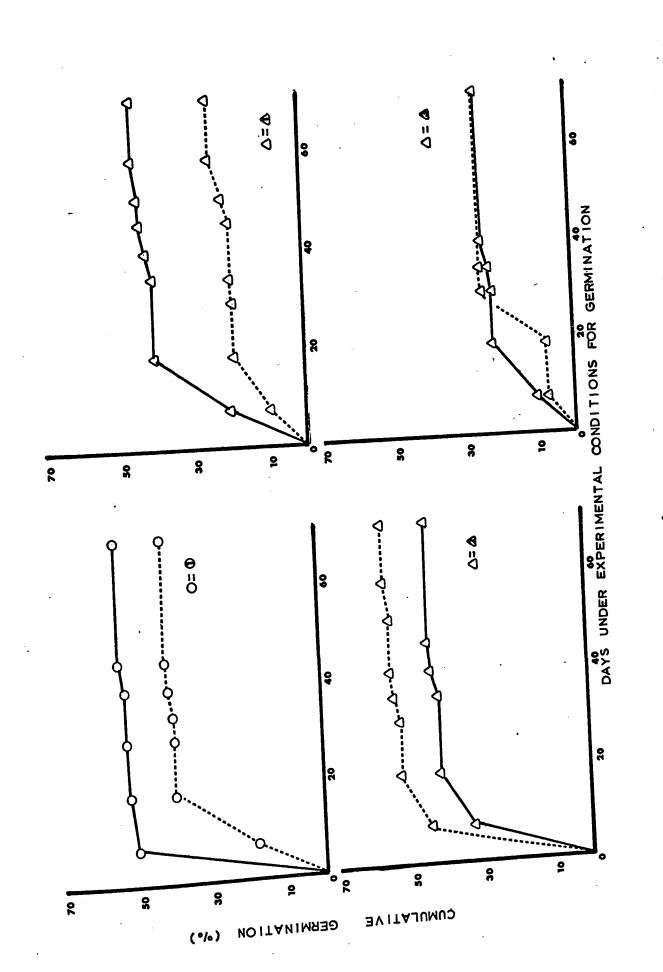
Figure 7.24A A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of collection R1 collected at two different harvests from plants of sowing one.

Figure 7.24B A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of collection P1 collected at two different harvests from plants of sowing one.

Figure 7.24C A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of collection P2 collected at two different harvests from plants of sowing one.

Figure 7.24D A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of collection P3 collected at two different harvests from plants of sowing one.

Notes: 1 - In each of these figures germination is expressed as a percentage of the total number of seeds sown.





- Figure 7.25A A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection R1 collected at two different harvests from plants of scuing one.
- Figure 7.25B A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection P1 collected at two different harvests from plants of sowing one.
- Figure 7.25C A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection P2 collected at two different harvests from plants of sowing one.
- Figure 7.25D A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection P3 collected at two different harvests from plants of sowing one.
- Notes: 1 In each of these figures germination is expressed as a percentage of the total number of seeds sown.
  - 2 In order to interpret these figures consult table 7.10 for a key to the symbols used.

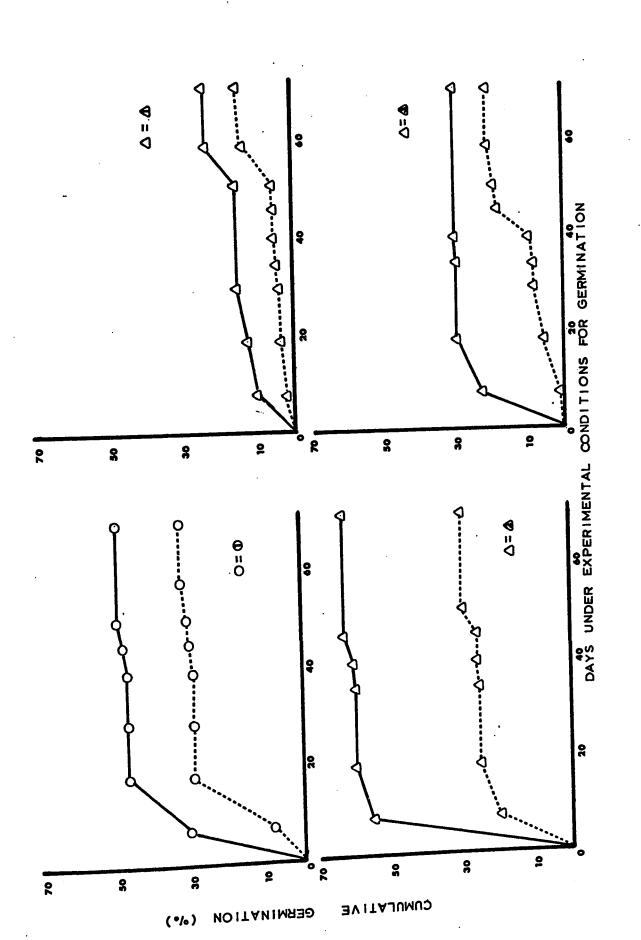




Figure 7.26A A comparison of the germination (in the field), after wintering in two different situations, of seeds of collection R1 collected at harvest one from plants of sowing two.

Figure 7.26B A comparison of the germination (in the field), after wintering in two different situations, of seeds of collection P3 collected at harvest one from plants of sowing two.

Figure 7.26C A comparison of the germination (in the field), after wint ring beneath the soil surface, of seeds of collection P2 collected at two different harvests from plants of sowing three.

Figure 7.26D A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection P2 collected at two different harvests from plants of sowing three.

Notes: 1 - In each of these figures germination is expressed as a percentage of the total number of seeds sown.

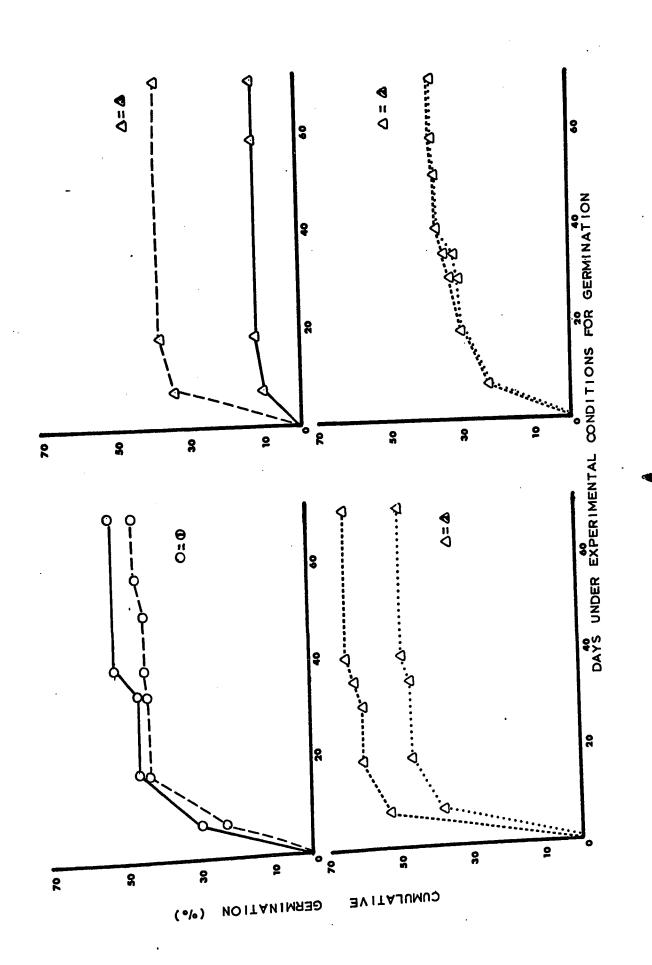




Figure 7.27A A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of collection P3 collected at two different harvests from plants of sowing three.

Figure 7.27B A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection P3 collected at two different harvests from plants of sowing three.

Figure 7.270 A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of collection R1 collected at two different harvests from plants of sowing three.

Figure 7.27D A comparison of the germination (in the field), after wintering on the soil surface, of seeds of collection R1 collected at two different harvests from plants of sowing three.

Notes: 1. - In each of these figures germination is expressed as a percentage of the total number of seeds sown.

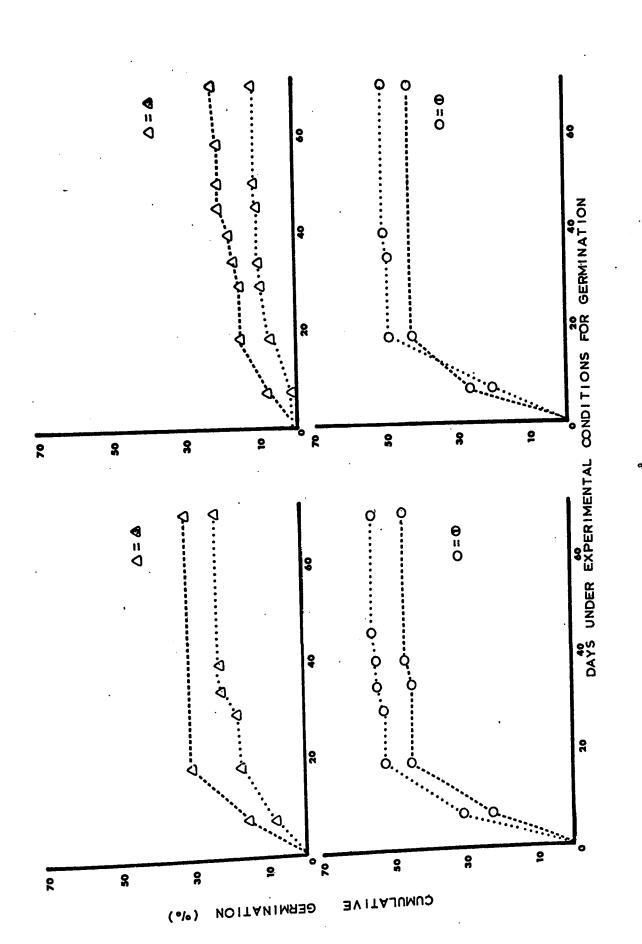




Figure 7.28A A comparison of the germination (in the field), after wintering in two different situations, of seeds of collection P2 collected at harvest one from plants of sowing one.

Figure 7.28B A comparison of the germination (in the field), after wintering in three different situations, of seeds of collection P2 collected at harvest two from plants of sowing one.

Figure 7.280 A comparison of the germination (in the field), after wintering in three different situations, of seeds of collection P2 collected at harvest two from plants of sowing three.

Figure 7.28D A comparison of the germination (in the field), after wintering in three different situations, of seeds of collection P2 collected at harvest three from plants of sowing three.

- Notes: 1 In each of these figures germination is expressed as a percentage of the total number of seeds sown.
  - 2 In order to interprest these figures consult table 7.10 for a key to the symbols used.

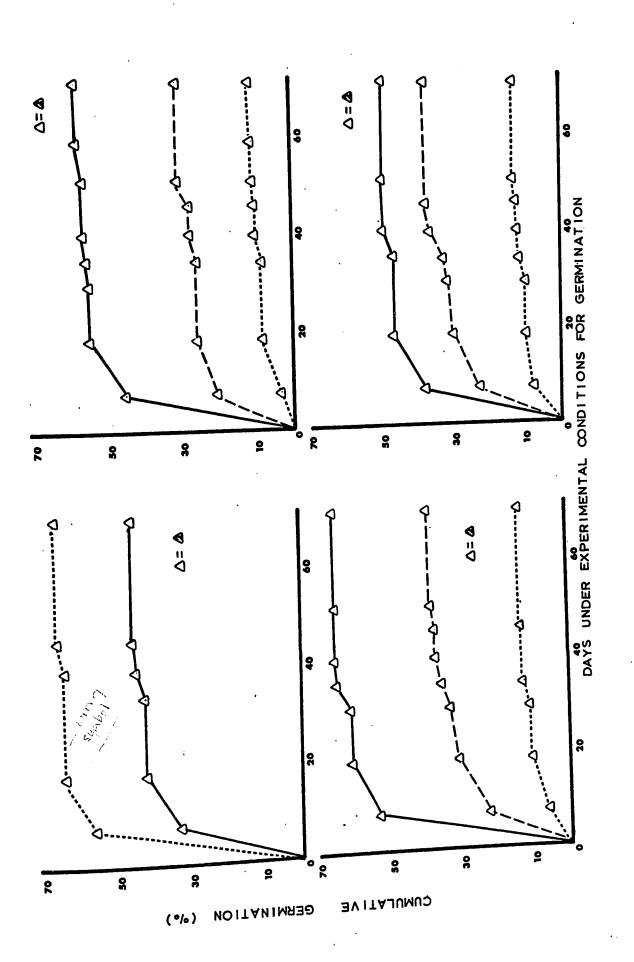




Figure 7.29A A comparison of the germination (in the field), after wintering in two different situations, of seeds of collection P3 collected at harvest one from plants of sowing one.

Figure 7.29B A comparison of the germination (in the field), after wintering in three different situations, of seeds of collection F3 collected at harvest two from plants of sowing one.

Figure 7.29C A comparison of the germination (in the field), after wintering in three different situations, of seeds of collection P3 collected at harvest two from plants of sowing three.

Figure 7.29D A comparison of the germination (in the field), after wintering in three different situations, of seeds of collection P3 collected at harvest three from plants of sowing three.

Notes: 1 - In each of these figures germination is expressed as a percentage of the total number of seeds sown.

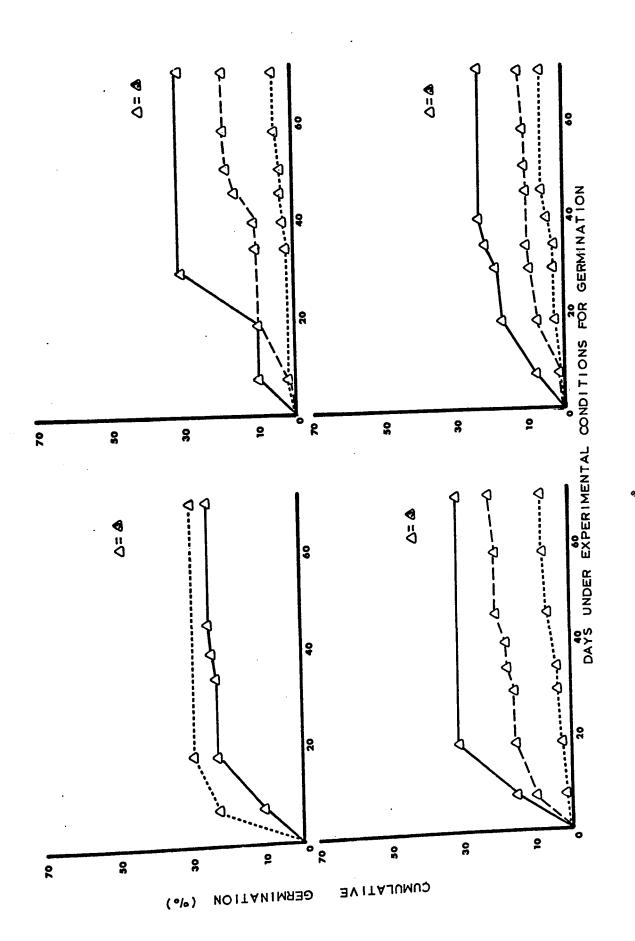




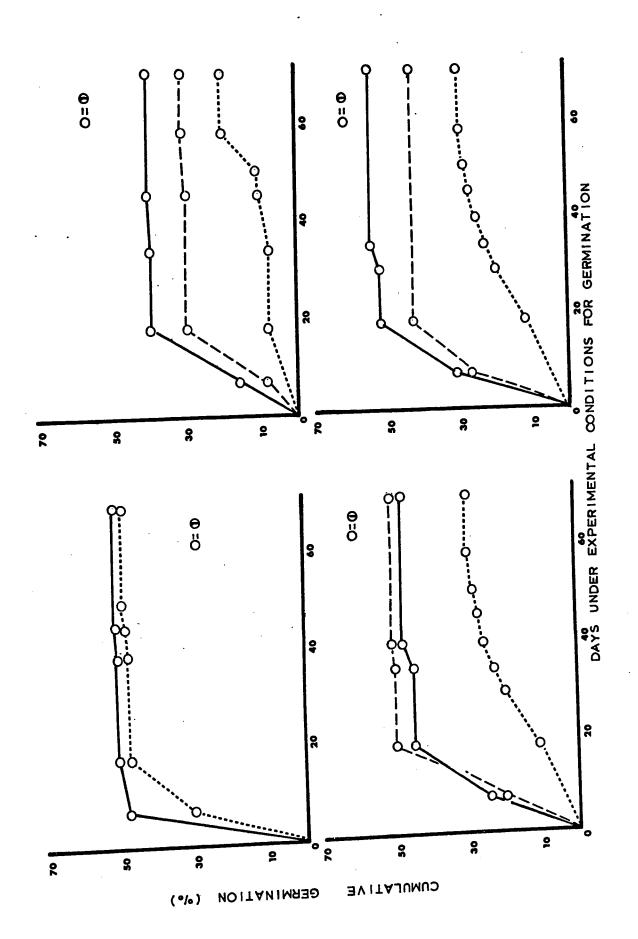
Figure 7.30A A comparison of the germination (in the field), after wintering in two different situations, of seeds of collection R1 collected at harvest one from plants of sowing one.

Figure 7.30B A comparison of the germination (in the field), after wintering in three different situations, of seeds of collection R1 collected at harvest two from plants of sowing one.

Figure 7.30C A comparison of the germination (in the field), after wintering in three different situations, of seeds of collection R1 collected at harvest two from plants of sowing three.

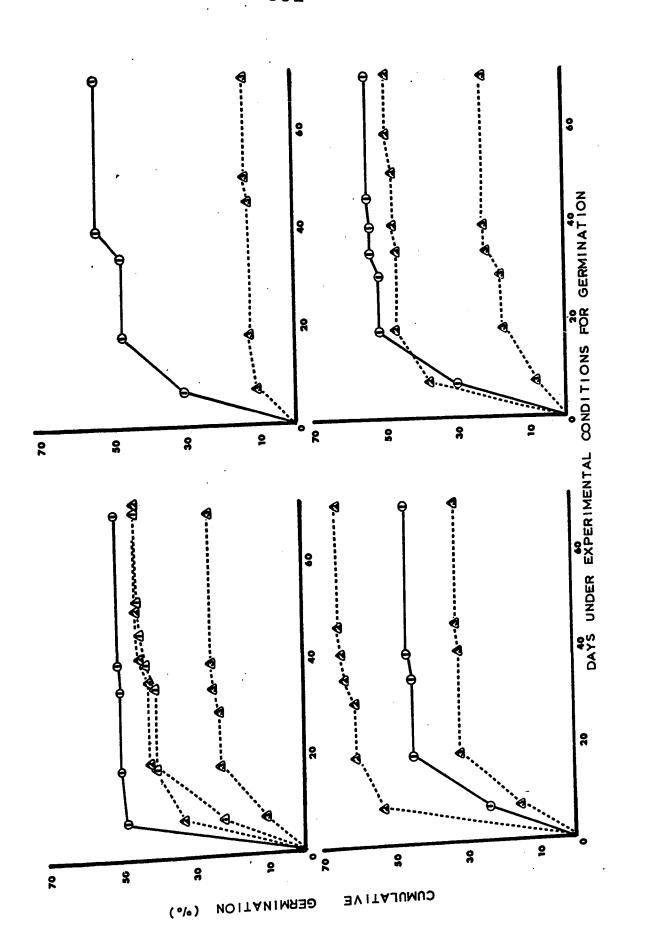
Figure 7.30D A comparison of the germination (in the field), after wintering in three different situations, of seeds of collection R1 collected at harvest three from plants of sowing three.

Notes: 1 - In each of these figures germination is expressed as a percentage of the total number of seeds sown.

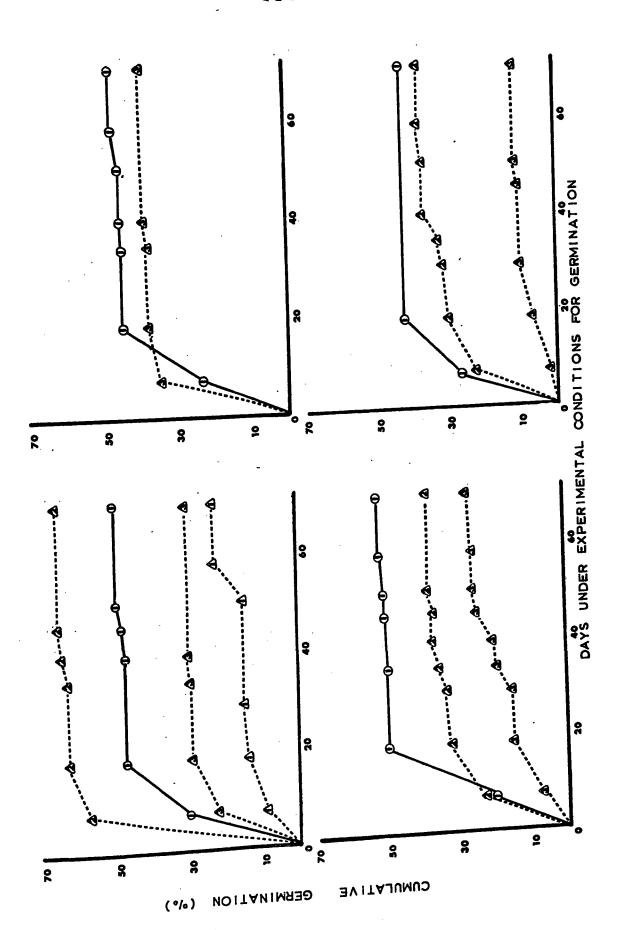




- Figure 7.31A A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of four different collections collected at harvest one from plants of sowing one.
- Figure 7.31B A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of three different collections collected at harvest one from plants of sowing two.
- Figure 7.31C A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of three different collections collected at harvest two from plants of sowing three.
- Figure 7.31D A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of three different collections collected at harvest three from plants of sowing three.
- Notes: 1 In each of these figures germination is expressed as a percentage of the total number of seeds sown.
  - 2 In order to interpret these figures consult table 7.10 for a key to the symbols used.

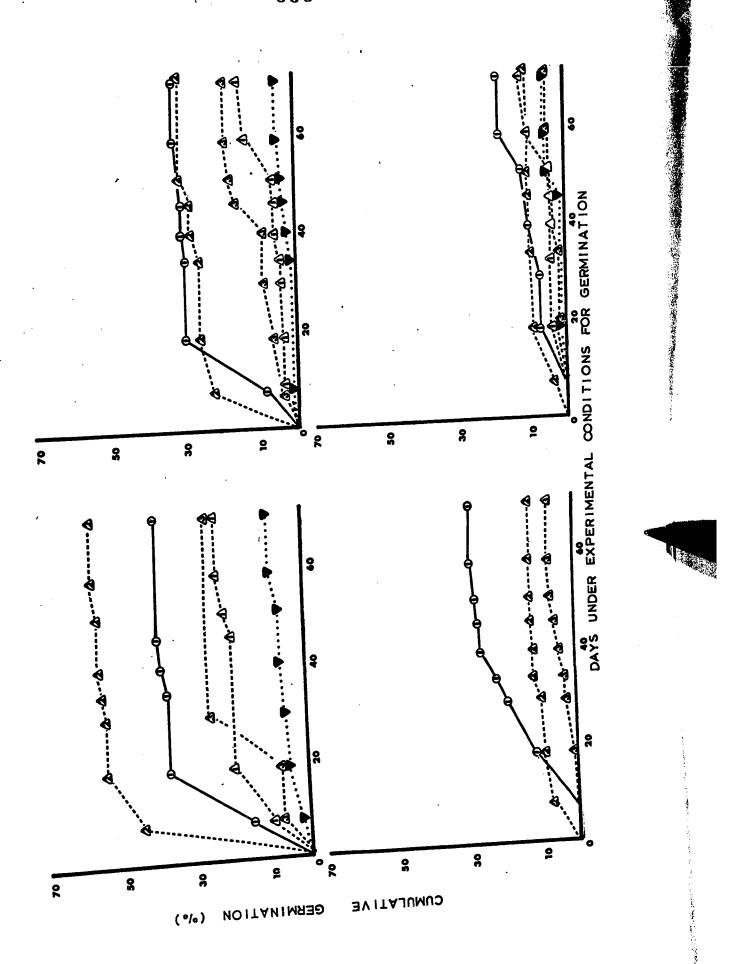


- Figure 7.32A A comparison of the germination (in the field), after wintering on the soil surface, of seeds of four different collections collected at harvest one from plants of sowing one.
- Figure 7.32B A comparison of the germination (in the field), after wintering on the soil surface, of seeds of three different collections collected at harvest one from plants of sowing two.
- Figure 7.320 A companison of the germination (in the field), after wintering on the soil surface, of seeds of three different collections collected at harvest two from plants of sowing three.
- Figure 7.32D A comparison of the germination (in the field), after wintering on the soil surface, of seeds of three different collections collected at harvest three from plants of sowing three.
- <u>Motes:</u> 1 In each of these figures germination is expressed as a percentage of the total number of seeds sown.
  - 2 In order to interpret these figures consult table 7.10 for a key to the symbols used.





- Figure 7.33A A comparison of the germination (in the field), after wintering beneath the soil surface, of seeds of <u>five different</u> collections collected at harvest two from plants of sowing one.
- Figure 7.33B A comparison of the germination (in the field), after wintering on the soil surface, of seeds of five different collections collected at harvest two from plants of sowing one.
- Figure 7.33C A comparison of the germination (in the field), after wintering on plant remains, of seeds of **three** different collections collected from plants of sowing **three**.
- Figure 7.33D A comparison of the germination (in the field), after wintering on plant remains, of seeds of five different collections collected from plants of sowing one.
- Notes: 1 In each of these figures germination is expressed as a percentage of the total number of seeds sown.
  - 2 In order to interpret these figures consult table 7.10 for a key to the symbols used.



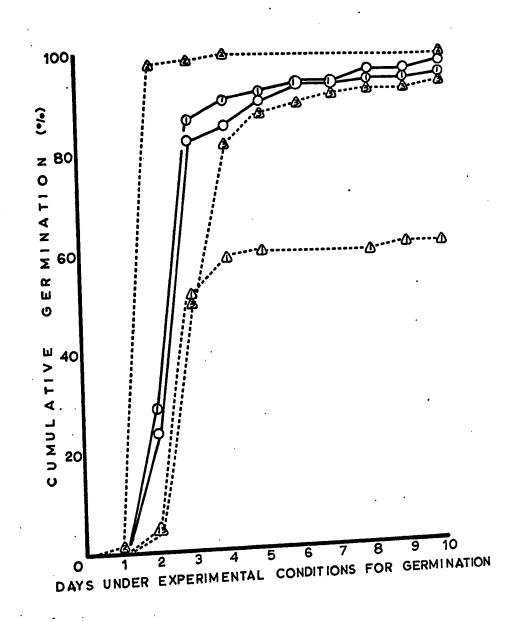
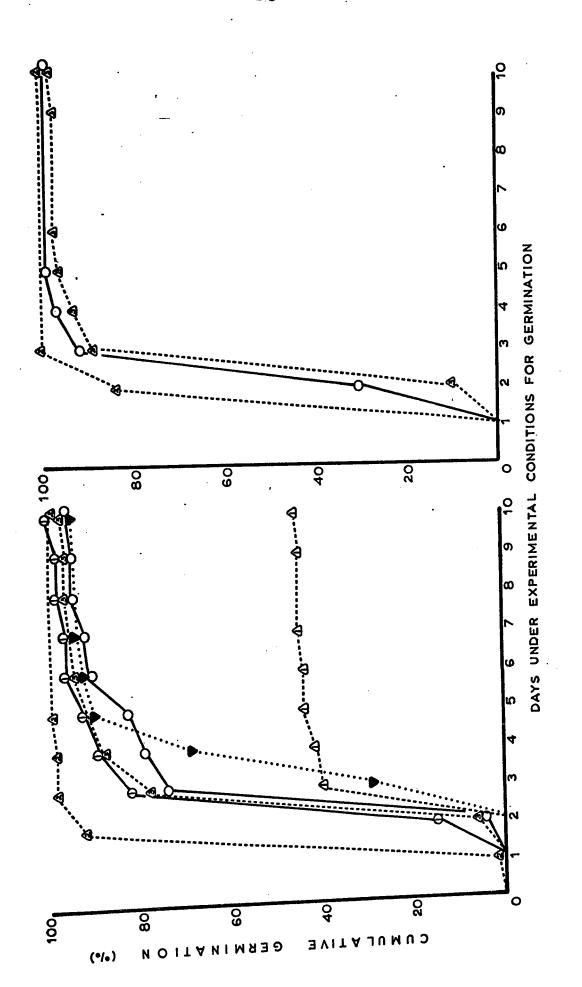


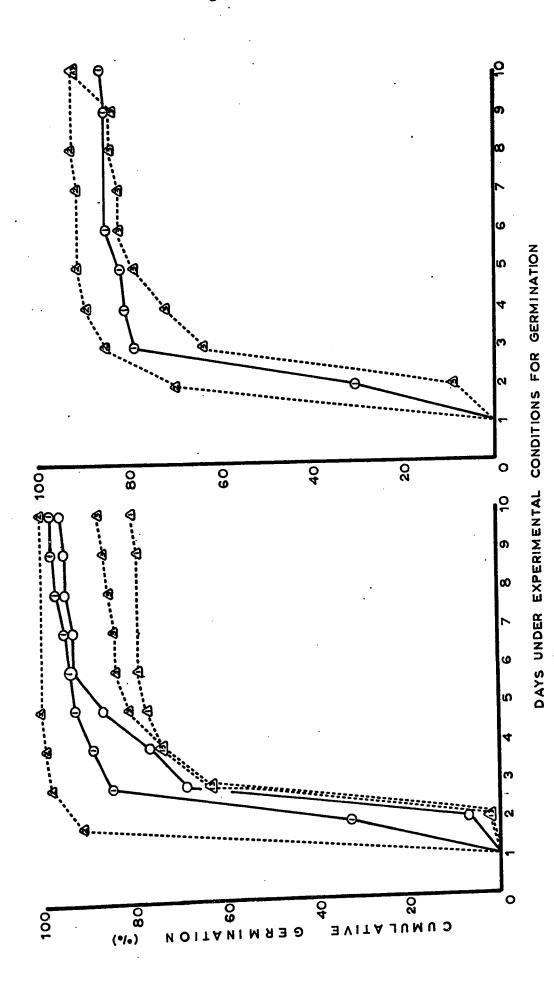
Fig. 7.34 The germination, after wintering for 17 months beneath the soil surface, of seeds of four different collections, collected at harvest one from plants of sowing one.

- Figure 7.35A A comparison of the germination, after wintering for 17 months beneath the soil surface, of seeds of five different collections, collected at harvest two from plants of sowing one.
- Figure 7.35B A comparison of the germination, after wintering for 17 months beneath the soil surface, of seeds of three different collections, collected at harvest two from plants of sowing three.
- Note: In order to interpret these figures consult table 7.10 for a key to the symbols used.



A comparison of the germination, after wintering for five months on the soil surface followed by twelve months beneath the soil surface, of seeds of <u>four different collections</u>, collected at harvest two from plants of sowing one.

A comparison of the germination, after wintering for five months on the soil surface followed by twelve months beneath the soil surface, of seeds of three different collections, collected at harvest two from plants of sowing three.



## ASPECTS OF THE COMPARATIVE BIOLOGY OF THREE WEEDY SPECIES OF ALARANTHUS IN SOUTHWESTERN ONTARIO

by

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## Vol. 11

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## 7.4 Experiment 10

The experiment was planned to investigate the germination behaviour of (a) seeds that had developed and overwintered on different parts of the plant, (b) seeds retained within utricles as compared with seeds which had been removed from their utricles, (c) seeds in alternating light and darkness, and seeds in continuous darkness, and (d) seeds exposed to a series of increases in temperature. A larger number of collections were available for inclusion in this experiment than in experiment 9 allowing a wider interpretation of the results in terms of local representatives of each species.

The seeds that were used in this investigation came from plants that had been grown under uniform field conditions during 1967 in order to provide specimens for the taxonomic study described in Chapter 3. When it was noticed in March, 1968 that the plants not used for herbarium specimens still retained a large number of seeds, it was decided to use these seeds in this experiment.

This experiment was begun before the second germination trial of experiment 9 and the results were used to determine some of the conditions that were to be employed in that experiment. The results of this experiment provide also a comparison with some of the results of experiment 9.

7.4.1 Materials and Methods

7.4.1.1 Culture of plant material under uniform conditions
Plants were grown from seed of the following species
and collections: P1, P2, P3, P5, P6, R1, R4, R5, and H7.

Details of the habitats and localities from which these collections were made is presented in table 6.2, page 212. The seed was sown and the seedlings cultured and transplanted to the field following the same procedures used in culturing plants for experiment 9 (described on page 243). Only one sowing was made in this experiment, seven days before the first sowing of experiment 9, on 3rd May, 1967. Each stage in the culture of the plants was approximately a week in advance of the first sowing of experiment 9.

Twenty five plants of each collection were planted out one metre apart in rows one metre apart. Those that were not used as specimens by the end of the growing season were left in the field over winter with no further treatment.

7.4.1.2 Collection of the seeds

The seeds were taken from the field on 31st March, 1968. The remains of entire inflorescences were detached from three plants (where possible) of each collection. The terminal inflorescence and the inflorescence of the lowest axillary branch were collected from each plant. Each inflorescence was placed in a separate paper bag and the bags were brought into the laboratory. After inflorescences had dried in the air the seeds were shaken free. The seeds to be used in germination trial 2 were placed in nylon bags and returned to the field where they were hung from a fence at a height of 1 metre above the soil surface until needed. 7.4.1.3 The assignment of seeds to treatments

As the seeds were harvested in the field they were assigned to categories that described (a) the position they

had occupied on the parent plant, (b) the individual plant from which they were taken, and (c) the collection and (d) the species to which the parent plant belonged.

It had been observed in the field that seeds that fell from the parent plant came to rest on the soil surface in one of two conditions; either they were still enclosed within the utricle or else the utricle had dehisced and separated from the seed. As seeds were shaken from the inflorescences they were separated into those that retained their utricles and those that had lost their utricles.

Seeds were assigned to different light regimes at the time the germination experiment was set up.

7.4.1.4 Treatment combinations

TABLE 7.11

A SYNOPSIS OF THE COMBINATIONS OF TREATMENTS EXAMINED IN EACH GERMINATION TRIAL IN EXPERIMENT 10

#### Germination trial 1 Light regime: Alternating light Constant darkness and darkness Position on Terminal basal Terminal basal plant: <sub>+</sub>2 Utricle: Collection Plant x<sup>1</sup> P1 1 $\mathbf{x}$ X X X $\mathbf{x}$ 2 $\mathbf{x}$ X 3 X $\mathbf{x}$ P2 1 X X X X. 2 X X x X

Light regime:			ernati nd dan		Light ss	Cons	stant	darl	cness
Position on plant:			minal	l bas	sal	Tei	rmina	l bas	sal
Utricle:		+2	-	+	_	+	-	+	-
Collection	Plant		<del></del>				- <del></del>		<del></del>
P3	1 2 3	x	x x	x	x x x		×		x
P5	1 2 3	x	x x	x	x x	x	x	x	x
P6	1 2 3	x	x x		x x		x		
R1	1 2 3	x x x	x	x x	x	x	x	x	x
<b>R</b> 4	<b>1</b> 2	x	x	x	x	x	x	x	x
R5	1	x	x	x	x	x	x	x	
Н7	1 2 3	x	x x	x	x x	x	x	x	x

# b) Germination trial 2

In the second germination trial only differences between collections were examined. Seeds from the terminal inflorescence of each plant were pooled for each collection. The following collections were included: P1, P2, P3, P4, P5, P6, R1, R4, R5, H7.

Notes: 1 - Those treatment combinations included are marked with an "x".

2 - + and - refer to seeds set to germinate with or without utricles respectively.

There was neither room nor time to examine the germination responses of seeds representing all possible combinations of factors. Those combinations that were included are

indicated in table 7.11. In order to determine which combinations were to be included, the following priorities were established: (a) It was more convenient to examine germination under alternating light and darkness than in continuous darkness. Therefore most comparisons were made under the former conditions. (b) An attempt was made to examine the germination response of seeds of all treatment combinations for one plant of each collection. For the remaining plants of each collection, the comparison between seeds with and without utricles was omitted. Other comparisons involving these plants were made using seed either with or without utricles depending on which was the most frequent condition in a particular collection. (c) The seeds of two collections of A. powellii shed their utricles It was not possible to collect sufficient very easily. seeds that retained their utricles to include all the planned treatment combinations for these collections.

#### 7.4.1.5 Germination trials

#### a) Trial 1: April, 1968 - laboratory

Three replicates of 50 seeds each of each treatment were counted onto Green's number 450 filter paper in 9 cm glass petri-dishes. For those treatments involving seeds retained within utricles, an additional five seeds (fruits) were included to allow for the inclusion of fruits with inviable seeds. This addition was necessary since only those seeds that appeared to be sound were included in those treatments in which the utricles were removed from the seeds. Ten millilitres of distilled water were added

to each petri-dish and the dishes were arranged on a table in growth room 37D.

Those treatments that were to be subjected to continuous darkness were placed in aluminum containers as described in experiment 5 (p. 216). The remaining treatments were arranged on the table in growth room 37D as described for experiment 9. The temperature cycles were the same as those used in experiment 9; 25°C by day and 10°C by night from the beginning of the experiment, (2nd April) for six days. The temperatures were increased by 50° to 30°C and 15°C for the next six days, then to 35°C and 20°C for the next five days and finally to 40°C and 25°C for the last seven days. Germination was scored daily and scedlings that had germinated were removed. Treatments in continuous darkness were scored for germination only three times during the trial, after 13, 19 and 25 days. On each of these occasions the canisters were taken to a dark room for counting. There they were opened and germination scored with the aid of illumination from an electric flashlight fitted with green filters.

After the germination trial was concluded, petri-dishes containing ungerminated seeds were removed to a bench in greenhouse 14. Here they were observed for further germination in order to estimate the viability of the seeds used in each treatment. A 0.1% solution of giberrellic acid was added to stimulate further germination, and seeds that did not respond to this treatment were nicked with a razor blade. Further germination that occurred after this

scarification was recorded and the total germination provided an estimate of viability.

#### b) Trial 2: May, 1968 - Field

Four replicates of 500 seeds each of each of collections P1, P2, P3, P4, P5, P6, R1, R4, R5, and H7 were prepared. The seeds of each replicate were scattered on the surface of bare soil in plots 50 cm by 50 cm, 3 m apart. treatments were assigned to plots in a random-block arrangement, and eight control plots were included. Each plot was covered with less than a quarter of an inch of sieved, sterilised Guelph loam soil. A fine spray of tap water was added to each plot from a hand watering can. The only water added subsequently came from natural precipitation. The plots were examined at weekly intervals from the 23rd May, (the date on which the experiment was begun) until 12th August. Germination was scored in the same way as in trial 3 of experiment 9 (described on page 22). No final estimate was made of the presence in each plot of ungerminated but viable seeds.

#### 7.4.2 Results

The cumulative germination scores of seeds of each treatment in each germination trial are presented in Appendix 3, tables A3.13 and A3.14.

7.4.2.1 Germination trial 1 - April, 1968.

### a) General

Since it was not possible to examine the germination of seeds subjected to every possible combination of treatments, the results constituted an unbalanced design which

complicated their analysis. As a result, the four different comparisons that follow were analysed. The methods of analysis followed those outlined in Appendix 4.

# i) Comparison 10/1 - The effects of the position of seeds on the parent plant

The germination results were compared for seeds from pairs of treatments that included one treatment of seeds from a basal position and one treatment of seeds from a terminal position. In other respects the members of each pair were identical. A two-factorial analysis was performed using the accumulated daily percentages of germination under alternating light and darkness.

# ii) Comparison 10/2. - The factorial effects of the position of seeds, the state of the utricle and the collection

The germination results were compared for seeds from one plant each of seven collections from basal and terminal inflorescences both with and without utricles. A three-factorial analysis was performed using the accumulated daily percentages of germination under alternating light and darkness.

# iii) Comparison 10/3 - Differences in germination between plants, collections and species

The germination results were compared for seeds from difference plants of the same collection, from different collections of the same species, and from different species. The analysis was performed initially in two stages. In the first stage data for seeds from terminal inflorescences were included and in the second stage data for seeds from

basal inflorescences were included. The state of the utricle varied between collections depending upon the state in which the majority of seeds of a collection were dispersed. A hierachical analysis of variance (described in Appendix 4) was performed using the accumulated daily percentages of germination of seeds in alternating light and darkness.

# iv) Comparison 10/4 - The effects of light during germination

The germination results were compared for seeds of different collections set to germinate either in alternating light and darkness or in continuous darkness. The analysis was performed in two stages. In the first stage results for seeds from basal and terminal inflorescences were compared from six collections. In the second stage only the results for seeds from terminal inflorescences were included and these were compared from nine collections.

Accumulated percentages of germination for days 13, 19 and 25 were analysed in a three-factorial design in the first stage of the comparison and in a two-factorial design in the second stage.

The analyses of these comparisons followed the methods described in Appendix 4. Details of the results of the analyses are presented in Appendix 5, beginning on page 609.

# b) Results of the statistical analyses

# i) Comparison 10/1 - Seed position

The analysis of variance of this comparison indicated significant differences between seeds from different positions.

It also revealed a significant interaction between seed position and the other treatments considered. Since these treatments were not logically related in this comparison, an investigation of the nature of this interaction was not performed. The examination of the effect of seed position was continued in comparison 10/2.

#### ii) Comparison 10/2

#### ii1) Differences between seeds from different positions

For most collections there was no significant difference in germination response between seeds taken from different positions on the same parent plant. However, seeds of collections P1 and R5 taken from basal inflorescences gave significantly greater initial germination than seeds from terminal inflorescences. The magnitude of these differences were small (compare Fig. 7.37 with 7.38 and compare Fig. 7.39 with 7.40).

# ii2) The influence of the utricle upon germination

For most collections there was no significant difference in germination response between seeds enclosed within utricles and seeds removed from their utricles. In collections P1, P3, R4 there was a greater cumulative germination towards the end of the trial for seeds retained within utricles than for seeds removed from utricles.

# ii3) Differences between collections

Many differences in germination response were observed between the seeds of different collections. These differences are presented in figures 7.37 to 7.40 and in

tables A5.29. The relationships between the germination of seeds of different collections were similar regardless of the position of the seeds on the parent plant and of the presence or absence of the utricle.

#### iii) Comparison 10/3

Hierachical analyses of variance of the cumulative germination scores of seeds from both terminal and basal positions revealed significant differences within species (i.e. between collections) and within collections (i.e. between plants of the same collection). No differences in germination were observed between species. In order to investigate further the nature of these differences, each of the two stages in the analysis was further divided into two components; (a) those treatments belonging to A. powellii, and (b) those belonging to A. retroflexus. Treatments belonging to A. hybridus were not considered further since this species was represented by only one collection. Details of the further analysis of these comparisons are presented in Appendix 5, page 620. The results of the revised analyses are discussed in the following paragraphs.

# iii1) <u>Differences between plants</u>

Significant differences in germination behaviour were observed between seeds taken from different plants of the same collection. These differences were pronounced among the collections of <u>A. powellii</u> but almost absent among collections of <u>A. retroflexus</u>.

#### iii2) Differences between collections

Significant differences in germination behaviour were observed between different collections of both <u>A. powellii</u> and <u>A. retroflexus</u>. These differences are presented in figures 7.37 to 7.40.

#### iii3) Differences between species

No differences in germination response were observed that distinguished species as a whole.

#### iv) Comparison 10/4

#### iv1) Differences in response to germination environment

A comparison of figures 7.37 and 7.39 with figure 7.41 illustrates the differences in germination response between seeds placed in continuous darkness and those placed in conditions of alternating light and darkness. Most treatments showed a significantly higher germination in alternating light and darkness than in continuous darkness during the early part of the experiment when temperatures were lower.

# iv2) Differences between collections

Significant differences in germination response were observed between the different collections examined under both germination conditions. Table A5.36 (Appendix 5) presents a summary of these differences.

# iv3) Differences between seed position

No significant differences were revealed between the germination responses of seeds collected from terminal and basal inflorescences in this comparison.

### 7.4.2.2 Germination trial 2 - May, 1968

#### a) Statistical analysis

The germination behaviour of seeds from different collections and species was examined in this germination trial, which was conducted in the field. The data were accumulated as totals of germination for each day on which observations were made, expressed as percentages of the final total in each plot. A comparison of the data provided information concerning the rates of germination but not concerning the absolute amount of germination.

The analysis took the form of an unbalanced hierachical design. Details of the results of the analysis are presented in Appendix 5, table A5.39.

### i) <u>Differences between collections</u>

Significant differences in percentage germination were observed between collections of the same species from the fourth to sixth days on which observations were made.

Inspection of the means in figure 7.42 suggests that collections differed more within A. retroflexus than within A. powellii. The striking deviation of collection P6 from other collections of A. powellii probably is not significant, since the ultimate number of seeds of this collection that germinated was very small (see table A3.14).

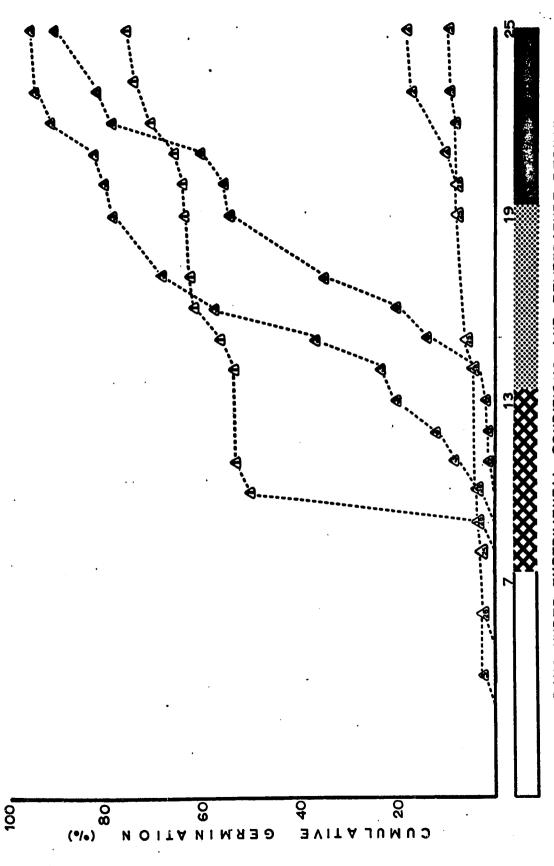
# ii) Differences between species

When the mean percentages of germination are considered for each species, it is seen that seeds of <u>A. powellii</u> germinated more rapidly than those of either <u>A. hybridus</u> or <u>A. retroflexus</u>.

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Fig. 7.37 The germination in alternating light and darkness of seeds from terminal inflorescences of plants of five different collections of A. powellii (seeds removed from utricles).

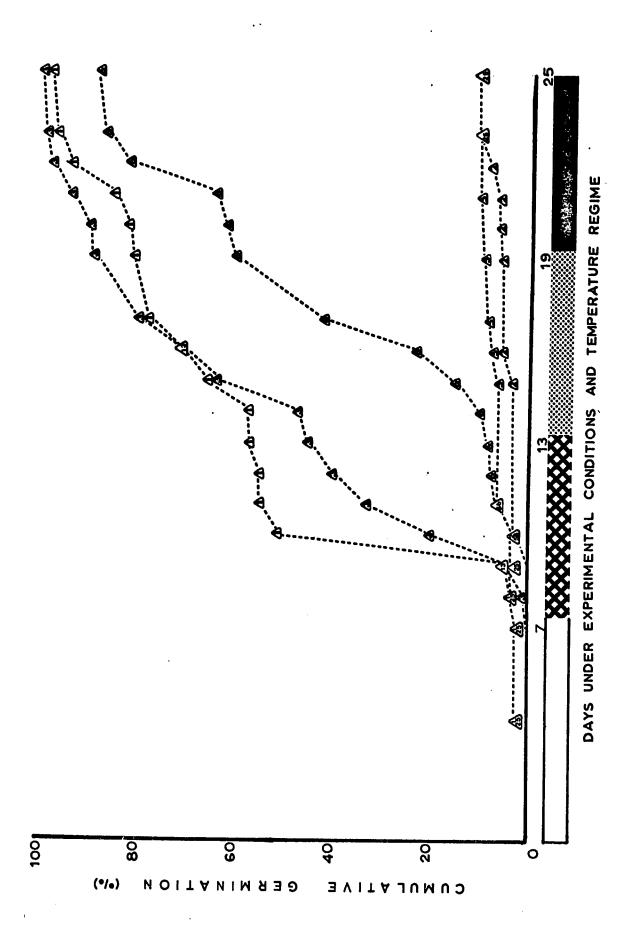
Mote: The symbols used in this figure are explained in table 7.10.



DAYS UNDER EXPERIMENTAL CONDITIONS AND TEMPERATURE REGIME

Fig. 7.38 The germination in alternating light and darkness of seeds from basal inflorescences of plants of five different collections of A. powellii (seeds removed from utricles).

Note: The symbols used in this figure are explained in table 7.10.



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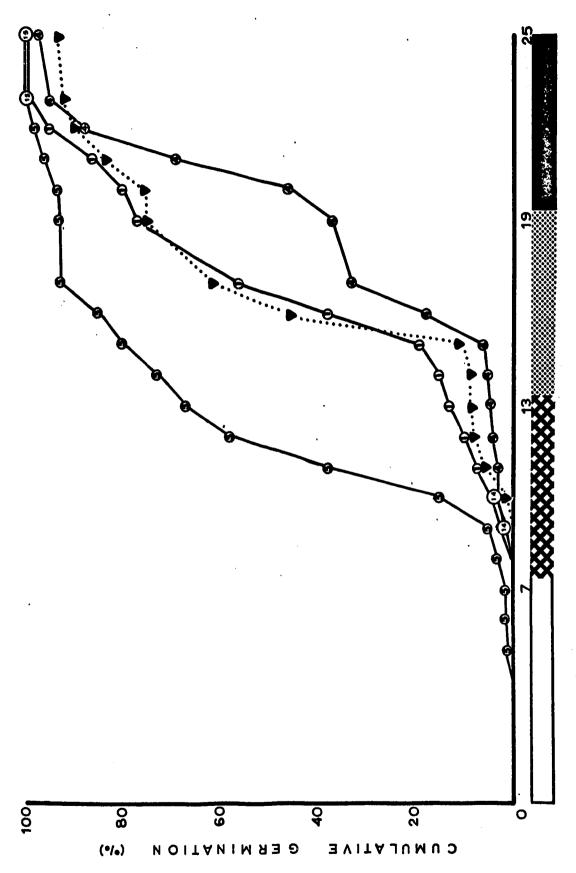
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Model Description of the second of the second

 $E_{\rm coll}(x,y,y,z) = e^{\frac{i\pi}{2} (x+y)^2} + 2E_{\rm coll}(y,y,z) + e^{-\frac{i\pi}{2} (x+y)^2} + 2E_{\rm coll}(y,z) + e^{-\frac{i\pi}{2} (x+y)^2} + e^{-\frac{i\pi}{2} (x+y)^2}$ 

The germination in alternating light and darkness of seeds from terminal inflorescences of plants of three different collections of A. retroflexus (seeds retained within utricles) and one collection of A. hybridus (seeds removed from utricles).

Note: The symbols used in this figure are explained in table 7.10.

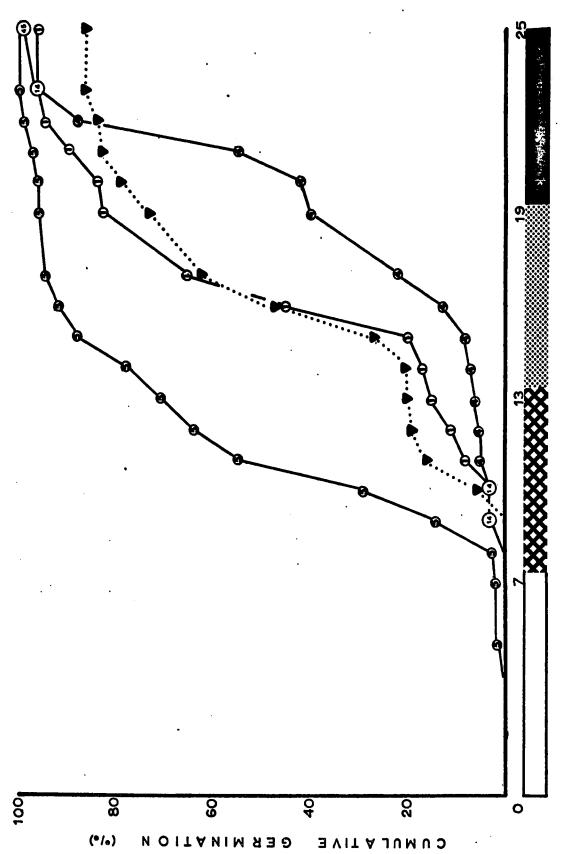


DAYS UNDER EXPERIMENTAL CONDITIONS AND TEMPERATURE REGIME

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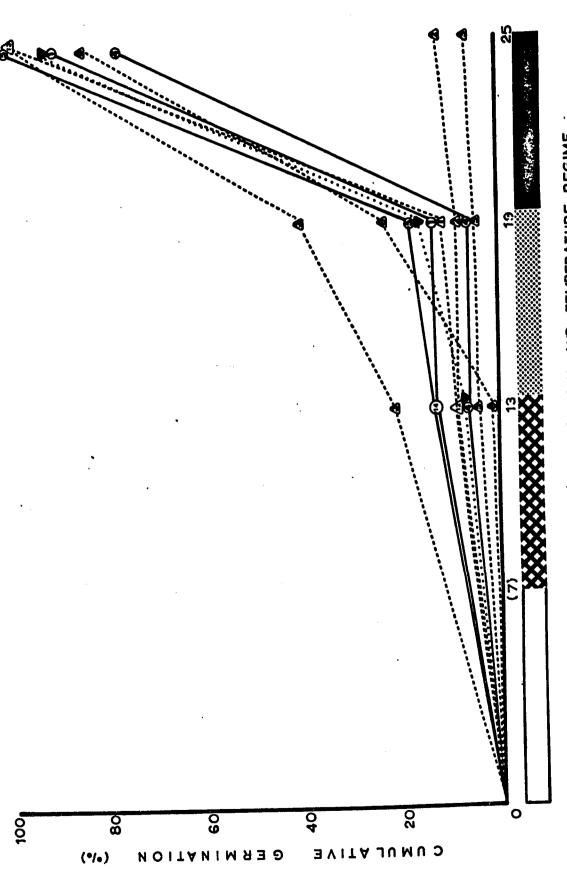
Fig. 7.40 The germination in alternating light and darkness of seeds from basal inflorescences of plants of three different collections of A. retroflexus (seeds retained within utricles) and one collection of A. hybridus (seeds removed from utricles).

Mote: The symbols used in this figure are explained in table 7.10.



DAYS UNDER EXPERIMENTAL CONDITIONS AND TEMPERATURE REGIME

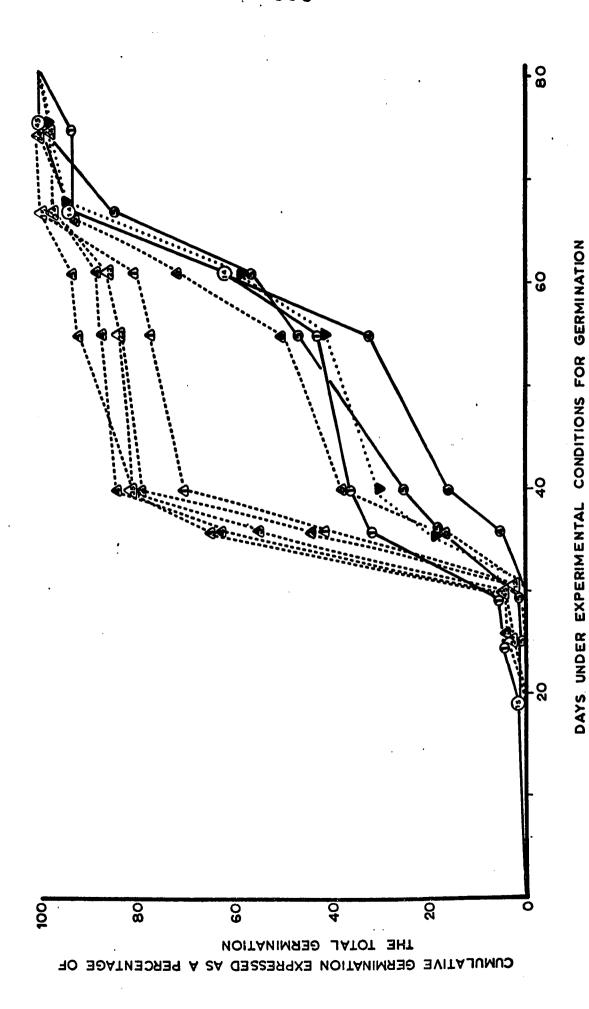
The germination in continuous darkness of seeds from terminal inflorescences of plants of one collection of <u>A. hybridus</u>, five collections of <u>A. powellii</u> and three collections of <u>A. retroflexus</u> (seeds removed from utricles).



DAYS UNDER EXPERIMENTAL CONDITIONS AND TEMPERATURE REGIME

Fig. 7.42 The germination in the field of seeds from plants of one collection of A. hybridus, six collections of A. powellii and three collections of A. retroflexus.

Note: The symbols used in this figure are explained in table 7.10.



7.4.3 Discussion of the results of experiment 10

It was pointed out in section 7.3.3 that in order to discuss any differences in the percentage of germination between seeds subjected to different treatments, it was first necessary to determine how the treatments themselves varied. In table 7.12 the components of variation contributing to each treatment in experiment 10 have been listed. These will be discussed in the following paragraphs.

TABLE 7.12

COMPONENTS OF VARIATION CONTRIBUTING TO EACH OF THE TREATMENTS INCLUDED IN EXPERIMENT 10

Levels of the factor included (Treatments)		ົ( ຣ	e ]	key	be	low	r)
P1 P2 P3 P4 P5 P6 R1 R4 R5	abcdef ghij	abcdef Shij	aaaaabbbc				
1 2 3	a b c						
terminal basal				a b	a b		
present absent						".	a b
alternating light and darkness continuous darkness						a b	
	factor included (Treatments)  P1 P2 P3 P4 P5 P6 R1 R4 R5 H7  1 2 3 terminal basal present absent alternating light and darkness continuous	factor included (Treatments) Gp  P1 a P2 b P3 c P4 d P5 e P6 f R1 g R4 h R5 i H7 j  1 a 2 b 3 c  terminal basal  present absent  alternating light and darkness continuous	factor included (SG GP GC)  P1	factor included (see (Treatments) Gp Gc Gs  P1	factor included (see key (Treatments) Gp Gc Gs I  P1	factor included (see key be (Treatments) Gp Gc Gs I P  P1	factor included (See key below (Treatments) Gp Gc Gs I P L  P1

#### Notes:

- 1 Individual components do not vary between treatments of each of the above factors if they are represented by the same letter in each vertical column.
- 2 Key to abbreviations for components:
  - Gp genotypic differences between individual
     plants
  - Gc genotypic differences between different collections
  - Gs genotypic differences between different species
  - I Internal physiological conditions of the parent plant as they impinge upon seeds produced at different positions
  - P Environmental conditions as they impinge upon seeds produced at different positions on the parent plant and remaining at different positions on plant remains over winter

- L Differences in light conditions as they influence the germination of seeds
- U The influence of the presence of the utricle on the germination of seeds

# a) The influence of the position at which seeds were borne on the parent plant

The following variables may have influenced the germination behaviour of seeds collected from basal and terminal inflorescences.

- 1) Internal phsiological differences during seed formation (Table 7.12, component I)
- 2) Differences in external environmental conditions between seeds in the two positions during their maturation and wintering (Table 7.12, component P).

It is interesting that the germination behaviour of seeds from basal and terminal inflorescences was so similar when in experiment 9 seeds that had wintered on plant remains were very different in their germination behaviour from those

that had wintered on the soil surface. In experiment 10, most of the basal inflorescences from which seeds were taken were resting on the soil surface at the time they were collected and appeared to have been in this position for a month or more. Thus the major ways in which seeds from basal branches in experiment 10 differed from seeds that had overwintered on the soil surface in experiment 9 were:

(a) the time of harvest, and (b) the proximity of the decaying remains of parent plants. The two germination trials also differed in being conducted approximately one month apart.

In experiment 9, the germination behaviour of seeds that had overwintered in the laboratory was taken as evidence that differences in the time of harvest between seeds that had and had not overwintered on plant remains were not responsible for the differences in germination response of these seeds. Thus it appears that the presence of decaying plant remains must be an important factor in controlling the germination of seeds that are retained on the plant remains over winter. Internal physiological factors and external environmental factors appear to have very little influence on seed germination or else their influence is masked by the influence of plant remains.

# b) The influence of the utricle

The environments of seeds with and without utricles may differ in temperature, light, the availability of oxygen and other factors. In this experiment it appears that such

differences have only a small influence on the germination In most collections germination of the seeds of seeds. was not influenced by the presence or absence of utricles. In three collections, (P1, P3 and R4) seeds enclosed within utricles completed their germination more rapidly than seeds without utricles. A hypothesis can be suggested to account for this phenomenom based on one of the observations from In that experiment it was noted that seeds experiment 6. of collection P2 that had imbibed in alternating light and darkness remained largely dormant whereas all of the seeds that had imbibed in darkness germinated rapidly. Moreover, once seeds had imbibed in alternating light and darkness, their subsequent transfer to continuous darkness did not result in increased germination; they had acquired a secondary dormancy.

The explanation of the differences in this experiment (10) may also involve a secondary dormancy. It is possible that certain of the seeds that were removed from their utricles acquired a secondary dormancy, perhaps in response to the alternating light and darkness. This dormancy might have been maintained for a long time if the temperatures at which the experiment was conducted had not been periodically increased. It is suggested that raising the temperatures overcame the dormancy with the result that the ultimate levels of germination of seeds with and without utricles were the same.

An objection can be raised in that the collections

whose seeds acquired secondary dormancy did not include collection P2, which was the collection involved in experiment 6. However, seeds of collection P2 exhibited a strong dormancy in this experiment, regardless of the presence or absence of utricles, and thus were not in a state to demonstrate the acquisition of a secondary dormancy.

In this experiment, the utricle was examined for the influence it might contribute to the control of germination at the moment when all other factors were optimal for germination. Since all of the seeds had passed the winter within utricles it was not possible to examine the influence of the utricle during after-ripening. In the context of seeds that have wintered on plant remains this may not be important since the majority probably remain within utricles. However among the seeds that are dispersed from the parent plant there are doubtless many seeds that pass the winter within utricles and many that do not retain their utricles over winter.

# c) The influence of light conditions

When placed in continuous darkness, most of the seeds of most collections required higher temperatures (which were provided later in the trial) before they would germinate than most of the seeds placed in alternating light and darkness. It is possible that this behaviour prevents the germination of buried seeds, unless they are very close to the surface where they would experience the high temperatures necessary for germination. However, it should be remembered that in experiment 9 it appeared that seeds that had wintered

on plant remains had not completed their after-ripening (and it will be shown that the results of this experiment confirm this opinion). Thus although the seeds used in this experiment showed a different response to the temperature sequence, depending upon light conditions, it cannot be assumed that the same responses would be obtained with fully after-ripened seeds. In experiment 9 it was shown that seeds that had been buried were further advanced in their after-ripening than seeds that had been retained upon plant remains.

Vegis (1963, 1964) has shown that partially afterripened seeds may have different temperature requirements
for germination in light and in darkness (discussed on
page 184). However he includes Amaranthus spp. among those
in which the temperature range that will permit germination
is greater in darkness than in light. The results of this
experiment do not agree with his conclusion. The
disagreement may results from different behaviours in
continuous light and alternating light and darkness, although
Vegis does not state which of these two conditions were
employed.

# d) Differences in germination between species and collections

# i) Within A. powellii

In alternating light and darkness seeds of collections of A. powellii germinated in a similar sequence in each treatment. Seeds of collection P1 were the earliest to germinate and these were followed by seeds of collections P3

and P6. The ultimate percentages of germination of seeds of each of these three collections were not significantly different from each other. However, the final percentages of germination of the seeds of these collections were significantly higher than those of seeds of collections P2 and P5. Most seeds of the last mentioned collections did not germinate even at the highest temperatures examined.

In continuous darkness the order of germination among collections was slightly different than in alternating light and darkness. Seeds of collections P3 and P6 gave significantly higher germination after 12 and 19 days than seeds of collection P1. However these three collections gave almost complete germination by the end of the experiment whereas seeds of collections P2 and P5 remained mostly ungerminated.

# ii) Within A. retroflexus

Different collections of A. retroflexus exhibited differences in their germination responses although the variation was not as great as that among collections of A. powellii. The collections of A. retroflexus germinated in a sequence that was maintained regardless of the treatments to which the seeds were assigned. Seeds of collection R5 germinated significantly earlier than those of collections R1 and R4, and seeds of collection R1 germinated earlier (although not always significantly so) than seeds of collection. R4. The final germination totals of the three collections were not significantly different.

## iii) Between A. hybridus, A. powellii and A. retroflexus

As a result of the wide variation in germination behaviour between collections of each species (and particularly of A. powellii) there were no distinct differences in germination behaviour that characterised each In comparing the germination of collections of different species the following generalisations can be made. The first collections in which a substantial number of seeds germinated were collections P1, P3 and R5. At the time that this germination began the temperature conditions were 30°C by day and 15°C by night. Collections P6, R1 and H7 were the next to begin to germinate and by this time the temperatures had been raised by 35°C and 20°C. Collection R4 was the last to germinate substantially and by the time that this collection had begun to germinate the temperatures had been raised to the final level of 40°C and 25°C. After seven days at these final temperatures only 10% of the seeds of collections P2 and P5 had germinated.

With the exception of collections P2 and P5, the general trend observed in experiment 9, that seeds of A. powellii tend to germinate before those of A. hybridus and A. retroflexus, is confirmed by the results of experiment 10.

# iv) Comparison of the results of this experiment and earlier experiments

The germination responses of seeds in experiment 10 reflected several of the differences observed between collections in experiment 3. The order in which collections

of A. retroflexus germinated was the same in the two experiments. Seeds of collection P1 were the first of A. powellii to germinate in both experiments and seeds of collections P2 and P5 were mostly dormant at both times. Seeds of collections of A. retroflexus were the first to exhibit substantial germination in experiment 3 whereas in experiment 10 seeds of several collections of A. powellii (P1, P3, and P6) were the first to exhibit germination. This difference may reflect differences in the conditions under which the experiments were conducted or differences in the conditions to which the seeds were exposed before the experiments.

The germination response of seeds in experiment 10 in continuous darkness lagged behind that in alternating light and darkness and was probably influenced by the periodic increases in temperatures. Thus at any point in time (except at the highest temperatures) a greater number of seeds had germinated in alternating light and darkness than in continuous darkness. These responses correspond with the responses of seeds from collections P3, R1 and R5 in experiment 6. Differences in details of the responses between the two experiments probably reflect the different conditions of the experiments and different pre-treatments of the seeds.

In experiment 6, seeds of collection P2 gave rapid and complete germination in darkness but remained dormant in alternating light and darkness. This striking difference

was not observed in experiment 10, in which seeds of this collection remained dormant both in darkness and in alternating light and darkness. The lack of agreement between these two sets of results may be related to the initial temperature conditions in which the seeds imbibed. In experiment 6 these conditions were favourable (depending on light conditions) for seeds of all collections, whereas in experiment 10 few seeds of any collection germinated until the initial temperatures were increased by 50°. Thus in experiment 10 a secondary dormancy may have been induced in seeds of collection P2 in response to the low initial temperatures.

The results of experiment 10 (for seeds without utricles, from terminal inflorescences) can be compared with the results of experiment 9 for seeds that overwintered on plant remains. Figures 7.19A, 7.37 and 7.39 illustrate this comparison. The order in which the different collections began their germination was the same in the two experiments. However, there were differences in the details of the two sets of responses which probably reflect the continuation of after-ripening in the interval between the two experiments. In both experiments germination of seeds began when the temperatures were raised to 30°C by day and 15°C by night. However the percentage of seeds of collections P1, R1 and H7 that germinated during the time that these temperatures were maintained were less in experiment 10 (conducted in April) than in experiment 9 (conducted in May). temperatures were increased to 35°C and 20°C, the differences

in germination between the two experiments disappeared. These observations conform with the concept of after-ripening proposed by Vegis (1963, 1964); as seeds after-ripen they are able to germinate at progressively lower temperatures.

The percentages of seeds of each collection that germinated under field conditions in experiment 10 were about the same as those of seeds of the same collection under field conditions in experiment 9. The only collection that was noticeably different was collection R1 of which 3.8% of seeds germinated in experiment 10 compared with 18.2% in experiment 9. There is no obvious explanation for this disagreement. However, the similarity of responses between seeds of different collections in this experiment and the similarity of response between seeds of collection R1 of different treatments in experiment 9 suggest that differences in the conditions under which the two experiments were performed were responsible for the different responses.

#### 7.5 Experiment 11

This experiment was designed to examine the germination responses of seeds of each species on several of the different types of soil found in the vicinity of London.

Two alternative approaches were available in planning this experiment. The first approach was to conduct germination trials in the locality in which each of the different soils

each of the different soils to the Department of Botany
Experimental Farm and to attempt to reproduce the different soil environments within one locality. The latter approach was chosen for its convenience and because, as a result, a greater number of extraneous variables (mostly climatic) could be kept uniform between the treatments.

7.5.1 Haterials and methods

7.5.1.1 The collection and preparation of soil

Soil Survey Report maps (Canada Department of Agriculture) were used to locate areas of each of seven different soil types. Each area was visited during early May, 1968 and eight litres of top-soil of each type was removed and taken to the Experimental Farm. Each quantity of soil was sterilised at 180°F for 12 hours and stored in covered plastic buckets until needed. A small quantity of each soil was analysed to determine its textural composition (following the methods outlined in Appendix 2). Table 7.13 presents detailed information for each soil.

One hundred and twelve clay drainage tiles, 15 cm in diameter and 30 cm high were munk to within 2 cm of their rims in eight adjacent rows. Within each row the tiles were spaced 12 cm apart and the rows were arranged in pairs, one metre apart, the two rows within a pair 12 cm apart. Each tile was filled to within 2 cm of the rim with sterilised soil from one of the collections of soil. A randomised block design was used in which there were four replicates of each soil fo each of the three species and for a control.

TABLE 7.13

	Guelph losm	T]	765636	Liddlesex	greyish brown	fair to good	slightly acid to neutral	, ,	°.'O	17%	34%	30%	19%
DIFFERENT SOILS INCLUDED IN EXPERIMENT 11	Perth silt loam	PSL	690873	Hiddlesex	brown	fair to good	slightly acid to neutral	,	), ()	754	74%	52%	27%
	Haldimnad clay	HC	531327	Hiddlesex	grey to light brown	fair to good	acid		) <sup>6</sup> (0	10%	23%	47%	. 56%
	Burford gravelly loam	BG	778615	Middlesex	light brown	ದಿಂಂಡೆ	slightly acid to neutral		25%	4455	26%	5/547	5,61
	Brookston clay loam	BCL	520289	Elgin	dark grey	poor	nevtral		5,0	13%	25%	5755	25%
DESCRIPTIONS OF THE	Berrien sandy loam	BEL	524287	Elgin	dark brown	poor	moderately acid		%0	21%	65%	12%	2%
DESCRIP	Berrien sand	BES	530298		yellow- brown	good to	moderately acid		<i>5,6</i> 0	%ंगर्प	sand 45%	%6	2%
	Name of soil	Soil	Coure Coure	County	~	Natural	Acidity 1	Analysis	Gravel	Coerse sand		Silt	Clay

drainage, colour and acidity (Canada Department of The name of each soil and details of natural vere determined from Soil Survey Report naps Agriculture) į ~ Lotes

The six-digit number is a reference figure (to within 100 metres) of the Universal Transverse Mercator Grid. 1 S

#### 7.5.1.2 The seeds used

The seeds used in this experiment came from the same plants as seeds used in experiment 10. The seeds were harvested on 31st March, 1968 and were stored in the field until 15th May, 1968 under conditions described in section 7.4.1.2 for seeds used in experiment 10 trial 2. Seeds of the following collections were included in this experiment; A. hybridus, collection H7; A. powellii, collection P3; and  $\Lambda$ . retroflexus collection R1.

#### 7.5.1.3 Germination trial

Four replicates of 500 seeds each of each collection were prepared. Seeds were chosen irrespective of the presence or absence of the utricle. The seeds were scattered on the soil surface in each tile and a small amount of sterilised soil was sieved over each (to a depth of less than 5 mm). Each tile was immediately watered with a fine spray of tap water from a watering can. The only subsequent water that was added came from natural precipitation. Observations of germination were made on approximately every seventh day from 15th May (the date of sowing) until the 12th August. Germination was recorded in the same way as described for the field germination trial of experiment 9, on page 252 . No final estimate was made of the presence of ungerminated but viable seeds in each tile.

#### 7.5.2 Results

The cumulative germination scores of seeds of each treatment are presented in table A3.15 in Appendix 3.

#### a) Statistical analysis

A two-factorial analysis was performed using the accumulated germination totals for each day on which observations were made. The analysis followed the methods described in Appendix 4 and the details of the analysis are presented in Appendix 5 beginning on page 630.

## b) Results of the statistical analysis

#### i) : The influence of soil type

The differences between the germination response of seeds on different soils are illustrated in figures 7.43 and 7.44. Table A5.42 in Appendix 5 presents a summary of the significance of these differences. For each species, germination was earliest and greatest on Berrien sandy loam and Guelph loam. At the other extreme, germination was the latest and least on Haldimand clay.

# ii) <u>Differences between species</u>

Differences between the germination of seeds of each species are illustrated in figures 7.43 and 7.44. In general, seeds of A. powellii germinated earliest and gave the greatest percentage of germination on each soil, whereas seeds of A. hybridus were the latest to germinate and gave the least percentage of germination on each soil. Seeds of A. retroflexus were intermediate in their response to those of A. hybridus and A. powellii. Only on Haldimand clay were there no significant differences between the percentages of germination of seeds of the different species and on this soil the percentages of germination were generally very small.

Fig. 7.43 The germination of seeds of each of the three species in the field in tiles containing different soils (see also Fig. 7.44).

Note: The symbols used in this figure are explained in table 7.10.

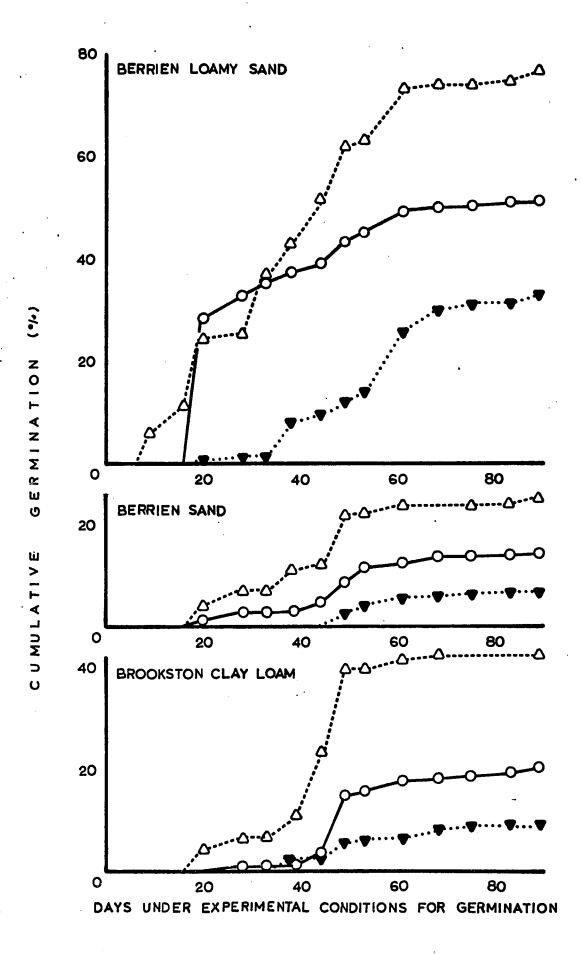
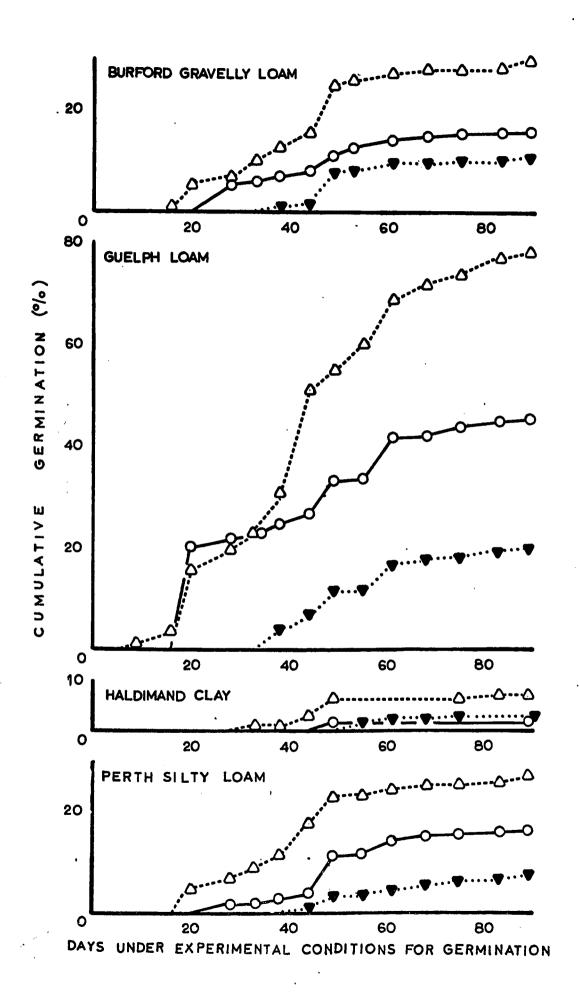


Fig. 7.44 The germination of seeds of each of the three species in the field in tiles containing different soils (see also Fig. 7.43).

Note: The symbols used in this figure are explained in table 7.10.



#### 7.5.3 Discussion of the results of experiment 11

In this experiment there were significant differences between the germination of seeds of different species on the same soil and between the germination of seeds of the same species on different soils. On almost every soil more seeds of A. powellii germinated than seeds of A. retroflexus and more seeds of A. retroflexus germinated than seeds of A. hybridus. Soils on which the greatest number of seeds of A. powellii germinated were also the soils on which the greatest number of seeds of A. hybridus and A. retroflexus germinated. Thus the influence of these two effects, soil type and species, appeared to be additive.

Two characteristics of each soil type that probably have influenced the germination response are colour and porosity. The colour of each soil, as described by the Soil Survey Report No.6 (Canada Department of Agriculture) is described in table 7.13. The appearance of the soils used agreed with these descriptions. No measurements were made of the porosity of the soils used but the following observations were made. The Berrien sand contained a high proportion of course sand. The surface of this soil dried rapidly after rain but the particles did not aggregate. In contrast, the surface of the Haldimand clay dried slowly after rain and tended to form a crust-like aggregation of particles when hot weather followed rain.

Ludwig and Harper (1958) reported that the temperature of the soil strongly influenced the time required for maize seedlings to emerge after sowing. They showed experimentally

that the surface colour of a soil had a strong influence on the daily temperature profile of the soil and the rapidity with which seedlings of maize emerged. Emergence was both greatest in number and the most rapid on dark-coloured soils and both the least in number and the slowest on light-coloured soils.

In this experiment germination was greatest (and earliest) on two of the darkest soils, Berrien sandy loam (described as "brown") and Guelph loam (described as "greyish brown"). The slowest and least germination was on Berrien sand ("yellow") and Haldimand clay ("grey to light brown"). Thus the germination patterns observed here, although for different types of soils, agree with those obtained by Ludwig and Harper (1958).

The texture of the soil may also influence its capacity to absorb heat. Daubenmire (1967) stated that coarse or well-aggregated soils respond more quickly to insolation than do heavy or poorly aggregated soils. He attributed this difference to differences in drainage and water content; wet soils are slower to warm up than dry soils. The soils on which the greatest germination occurred in this experiment tended to be those with good, but not excessive aggregation (e.g. Berrien sandy loam, Brookston clay loam, Guelph loam). The soils on which the greatest germination was observed in this experiment shared a combination of both of these characteristics that would favour the early warming of the soil.

#### 7.6 Experiment 12

#### 7.6.1 Introduction

This experiment was designed to determine to what extent seeds will germinate in the autumn immediately following their dispersal from the parent plant. The experiment was conducted under both field and laboratory conditions with seeds from plants of several different collections.

#### 7.6.2 Materials and methods

#### 7.6.2.1 Seeds used

At the end of **June**, 1963 one seedling in each plot of experiment 10 was allowed to grow to maturity. In this way two to five plants were provided of each of the following collections: P1, P2, P3, P4, P5, P6, R1, R4, R5 and H7. Seeds were taken only from those plants that were morphologically identical to plants of the original collections grown under uniform field conditions in 1967. 7.6.2.2 Collection of the seeds

The seeds were removed from the terminal inflorescence of each plant in the same manner as described in experiment 9 (section 7.3.2.2). Seeds were collected on 8th, 16th and 30th September. The seeds were separated from any bracts or pieces of inflorescence that were detached during collection. Seeds were used regardless of the presence or absence of the utricle.

#### 7.6.2.3 Germination trials

#### a) Germination trial 1 - field

One hundred and twenty of the flower pots used in experiment 9 (trial 3) were emptied of soil and refilled with sterilised soil. The pots were then replaced in the ground at the Department of Botany Experimental Farm.

Four replicates of 100 seeds each of each plant were used. The seeds were scattered on the surface of the soil in the pots. A randomised block design was employed. The seeds were sown on the day they had been harvested (8th September). Seedlings that had emerged were counted at weekly intervals until the beginning of November.

#### b) Germination trial 2 - laboratory

Four replicates **each of** 50 seeds were prepared from seeds of each collection harvested on 16th September. The seeds were counted onto Green's 450 filter paper in 9 cm glass petri-dishes and the petri-dishes distributed in a random block arrangement upon a table in growth room 370 (ss Table 6.3). The temperatures of the growth room were maintained at 35°C by day and 20°C by night; the daylength was 12 hours in each 24 hr cycle. Ten millilitres of distilled water were added to each petri-dish at the initiation of the experiment on 18th September and more water was added subsequently as required.

Observations of germination were made daily for ten days after which any ungerminated seeds were scarified with a razor blade. Later a final count of germination was made as an estimate of viability.

# c) Germination trial 3 - laboratory

Seeds collected on 30th September were pooled for all plants of the same collection. Six replicates each of 50 seeds of each of collections P1, P2, P3, R1 and H7 were counted onto Green's 450 filter paper in glass petri-dishes. Three replicates of each collection were placed in an incubator (see Table 6.3) maintained at 35°C during the 12 hr day and 20°C by night. The remaining three replicates were placed in an incubator (see Table 6.3) maintained at 30°C by day and 15°C by night, also under a 12 hr light/12 hr Within each incubator the replicates were dark cycle. Ten millilitres of arranged in a random block structure. distilled water were added to each dish on 1st October, the day the experiment was initiated, further water was added as Germination was scored in each incubator at necessary. daily intervals for eight days. At this time temperature conditions in the low temperature incubator were changed to 35°C by day and 20°C by night, and germination in these conditions was recorded for another seven days. completion of the trial all ungerminated seeds were scarified and further germination was noted to provide an estimate of viability.

#### 7.6.3 Results

#### a) Germination trial 1

Emerged seedlings were observed only at two of the times at which observations were made. The total number of seedlings that were observed was small and for this reason the results were not analysed statistically. Germination

scores are presented in table A3.16 in Appendix 3.

summary of the results is presented in table 7.14.

TABLE 7.14

THE MEAN TOTAL GERMINATION OF FRESHLY HARVESTED SEEDS SOWN IN POTS IN THE FIELD

Collection:	P1	P2	P3	P4	P5	P6*	R1	R4	R5	H7*
Plant 1	0	0	0	0	0	0	3.5	0.3	1.3	0
Plant 2	0	0	0	0.3	0	0	0.8	0	1.3	0
Plant 3	0	0	0	0.3		0	3.0	-	0	0

Notes: 1 - Values are mean percentages for four replicates of seeds from each plant.

# b) Germination trial 2

The seeds included in this experiment were maintained under uniform conditions in order to compare the responses of seeds from different plants, different collections and different species. Germination scores are presented in table A3.17, in Appendix 3. An unbalanced, nested analysis of variance was performed using the accumulated percentages of germination for each day of the trial. The details of the analysis are presented in tables A5.43 to A5.45 in Appendix 5.

The analysis revealed significant differences in the germination responses of the seeds between plants of the same collection, between collections of the same species and

<sup>2 -</sup> Seeds were sown 8th September, 1968 except for those collections marked with an asterisk (\*) seeds of which were sown on 16th September.

between different species. Duncan's multiple range test of the means for each species revealed that significantly more seeds of <u>A. powellii</u> and <u>A. retroflexus</u> germinated than seeds of <u>A. hybridus</u>. A summary of the mean values for the germination of seeds of each plant is presented in figure 7.45.

#### c) Germination trial 3

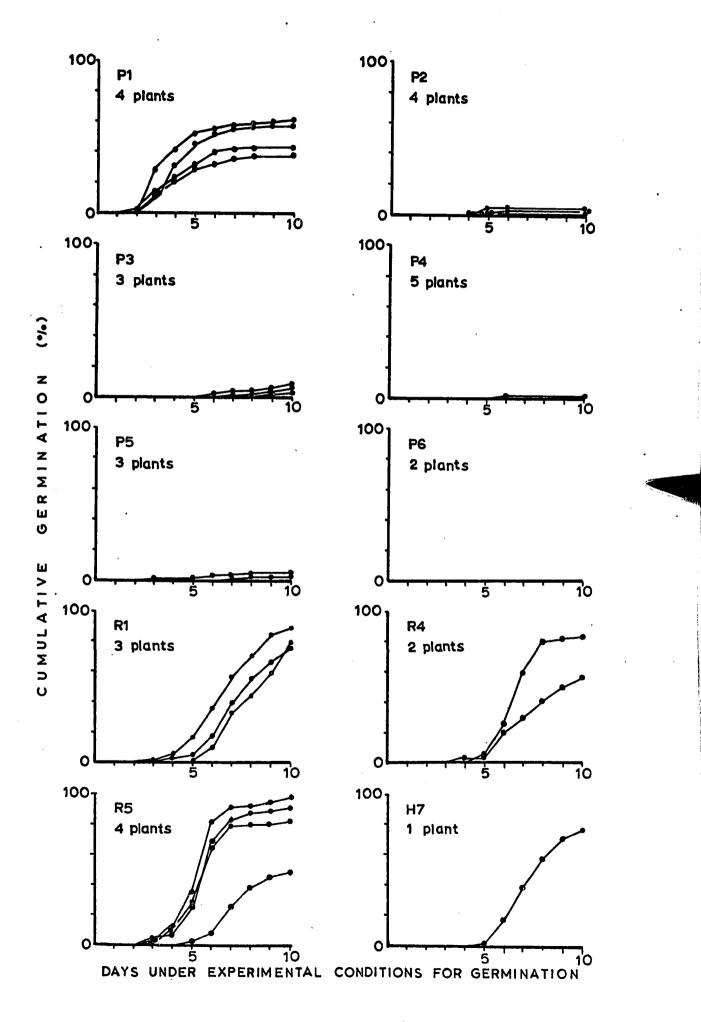
Germination scores for seeds of each treatment included in this trial are presented in table A3.18 in Appendix 3. The accumulated percentages of germination for each day of the trial were analysed by the method of two-factorial analysis of variance. A further analysis was made in order to compare germination totals after eight days at 35°C with germination totals after a combination of eight days at 30°C followed by seven days at 35°C. The purpose of this analysis was to determine whether seeds that imbibed at 30°C but did not germinate at that temperature differed in their response when the temperature was increased to 35°C from seeds that imbibed at 35°C. The complete details of these analyses are presented in Appendix 5 beginning on page 634.

# i) The influence of different temperatures

The percentage germination of seeds of each collection except collection P2 was significantly greater at the end of eight days at 35°C than at the end of eight days at 30°C.

Only a very few seeds of collection P2 germinated at either temperature. When seeds that remained ungerminated at the end of eight days at 30°C were subjected to a temperature of

Fig. 7.45 The germination of freshly harvested seeds of plants of collections of each species at alternating temperatures of 35° and 20°C in trial 2 of experiment 12.



35°C for seven days more seeds germinated and the ultimate percentage of germination was not significantly different from that which was observed in eight days at 35°C, for seeds of any one collection (Fig. 7.46).

# ii) Differences between collections

Few seeds of any collection germinated at 30°C, and thus no differences were observed between collections. Differences in the germination response of seeds of different collections subjected to 35°C are illustrated in figure 7.46. At this higher temperature significantly more seeds germinated of collections P1, R1 and H7 than of collections P2 and P3.

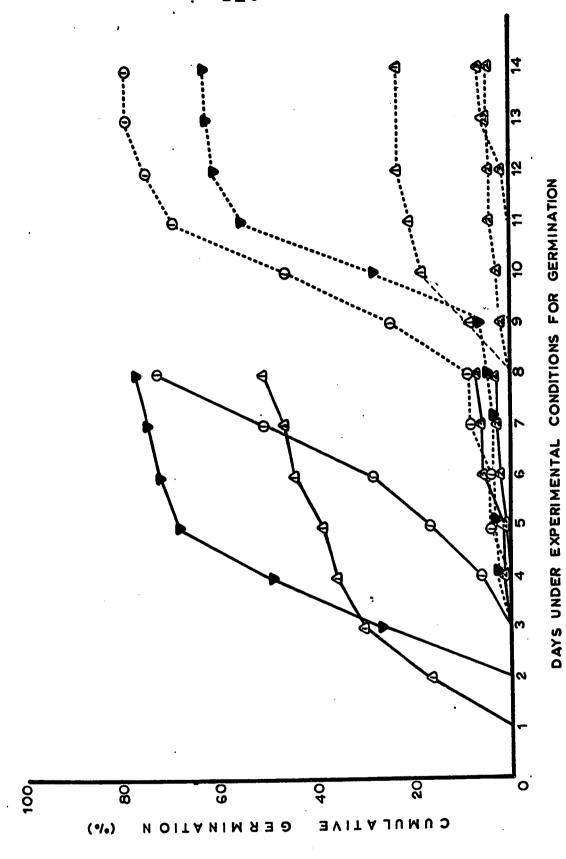
Differences in the germination responses of seeds of different collections subjected to 30°C followed by 35°C are presented in figure 7.46. Significantly more seeds germinated of collections R1 and H7 than of collections P2 and P3. The response of seeds of collection P1 was not significantly different form the response of seeds of any other collection.

### 7.6.4 Discussion

The high percentages of germination of seeds of some collections in the laboratory experiments contrasted with the low percentages of germination of seeds of the same collections in the field. This difference suggests that the temperatures maintained in the laboratory experiments were not encountered by seeds in the field, or were encountered for two short a period to stimulate germination.

Fig. 7.46 The germination of freshly harvested seeds of plants of different collections subjected to (a) alternating temperatures of 35° and 20°C for eight days (solid lines), or to (b) alternating temperatures of 30° and 15°C for eight days followed by seven days at 35° and 20°C (broken lines).

Note: The symbols used to identify the collections are described in table 7.10.



The different germination responses in the laboratory of seeds of different collections indicate that the seed production of plants of different collections differ in the proportions of seeds that are dormant under the particular conditions that were examined. Thus at 30°C, all of the seeds of collections of A. powellii were dormant whereas a small proportion of seeds of A. hybridus and A. retroflexus were non-dormant. A similar relationship between seeds of A. powellii and A. retroflexus was observed in the field trial. However, under these conditions, all seeds of A. hybridus were dormant.

#### 7.7 Experiment 13

#### 7.7.1 Introduction

This experiment was designed to investigate more precisely the influence of post-harvest conditions on the germination of seeds of A. powellii. In experiment 9, seeds of different collections of this species had exhibited different germination responses after various post-harvest treatments. In particular, seeds that had overwintered in contact with the soil (but not in contact with decaying plant remains) had responded differently in germination tests from seeds that had overwintered in the remains of the parent plant (and not in contact with the soil). It was hoped that in this experiment that it would be possible to identify more precisely the variable or variables responsible for the differences that were observed in germination in

experiment 9. Seeds of two collections of <u>A. powellii</u> were included in this experiment. The collections were chosen because of the dissimilarities in their responses in experiment 9.

7.7.2 Materials and methods

# 7.7.2.1 Seeds used

The seeds used in this experiment were collected from two of the plants of each of collections P2 and P3 that had provided seeds for experiment 12 (see section 7.6.2.1).
7.7.2.2 Collection of the seeds

The terminal inflorescences of secondary stems and tertiary stems (i.e. lateral branches and branches of lateral branches) were removed from the plants. The terminal inflorescence of primary stems was not collected since it was thought that seeds on this inflorescence would represent a wider range in ages than seeds from the remaining inflorescences.

# 7.7.2.3 Post-harvest treatments

# a) The presence of plant remains

In one treatment seeds were removed from an individual inflorescence and placed in a 12 cm by 3 cm nylon bag. In the other treatment an entire inflorescence bearing seeds was placed in a nylon bag.

# b) Wintering situations

Sixteen replicates of each of the above treatments were prepared for each collection, and placed in each of the five following positions: (i) 15 cm above the soil; (ii) 60 cm

above the soil; (iii) on the soil surface; (iv) 15 cm below the soil surface; and (v) 60 cm below the soil surface. For each of these treatments the bags containing the seeds were threaded onto a nylon line in such a way that the individual bags could be removed without breaking the line or opening the bags. The nylon lines carrying the aboveground treatments were supported by plastic-coated wire lines held at the appropriate height above the soil surface by wooden stakes. The nylon lines carrying the belowground and surface treatments were secured to wooden pegs (15 cm in length) which were then sunk into the ground. In each treatment the individual bags were separated by 15 cm on the nylon line.

# c) Length of the wintering period

The nylon bags containing the seeds were placed in the field on 8th November, 1968, five days after the collection of seeds. It was intended that four replicates of each treatment would be removed from the field on each of the following dates: 12th December, 1968, 21st January, 18th March and 18th April, 1969. However, it was not possible to retrieve bags from some of the surface and below-surface treatments on 21st January and 18th March because the soil was frozen. As a result a further retrieval of buried treatments was made on 3rd May, 1969.

# 7.7.2.4 Germination trials

Germination trials were conducted with freshly harvested seeds at the beginning of the experiment and with seeds that had wintered in the field at each of the times of retrieval.

# a) Initial germination trial

Immediately after the seeds were removed from the plants in November, 1968 a germination trial was conducted examining the response of seeds from four randomly chosen inflorescences of each collection. Four replicates of 50 seeds each were prepared from each inflorescence. The seeds were counted onto filter paper in glass petri-dishes and the petri-dishes were arranged in random blocks on the upper shelf of incubator 13/3. The incubator was maintained at 30°C by day and 15°C by night under a 14 hr day. Ten millilitres of distilled water were added at the beginning of the trial and germination was recorded daily for twelve days. At the end of this period viability was estimated after the seeds had been scarified.

# b) Germination after retrieval from field

At each of the retrieval times the bags containing seeds were brought into the laboratory, washed by immersing them gently in cold tap water and allowed to dry in the air for about an hour. Then the bags were opened and the seeds or inflorescence were removed. Where seeds had overwintered with remains of the inflorescence, only those seeds that remained lodged within the inflorescence were used in the germination trial. Fifty seeds from each of three bags of each treatment were counted into individual petri-dishes and placed in incubator 13/3 under the same conditions described for the pre-winter trial. Germination was scored at daily intervals for at least eleven days in each trial.

At the completion of each trial ungerminated seeds were scarified and subsequent germination was recorded as an estimate of viability.

### 7.7.3 Results

Cumulative germination scores for seeds of each treatment are presented in table A3. in Appendix 3.19 The results of each germination trial were analysed separately. Details of the analyses are presented in Appendix 5, beginning on page 637. The results of the initial germination trial were not analysed statistically, since the amount of germination was extremely low among seeds of each treatment and hence the results would not have been significant. The mean total percentages of germination after 11 days for seeds of each treatment are presented in figure 7.47.

# a) Influence of the wintering situation

At the first time of retrieval, differences were observed in the germination behaviour of seeds of collection P3 that had wintered without plant remains and in different positions relative to the soil surface. Those seeds that had wintered on the soil surface gave a significantly lower percentage of germination than seeds that had been suspended above the soil.

The most pronounced differences in germination behaviour that were related to wintering position were observed after the retrieval of seeds on 16th April. Seeds of collection P2 that had wintered without remains of the inflorescence gave much lower germination after wintering on the soil surface than after wintering in any other position. Seeds

- Fig. 7.47 Histograms illustrating the mean total germination after 11 days of seeds of each treatment in experiment 13
- Notes: 1 Histograms in the same column illustrate the results for seeds retrieved from the field at the same time. Histograms in the same row illustrate the results for seeds from the same post-harvest treatment.
  - 2 Within each histogram results are presented as four vertical bars which represent the following depending upon the pattern in which they are presented:

Collection P2

Collection P3

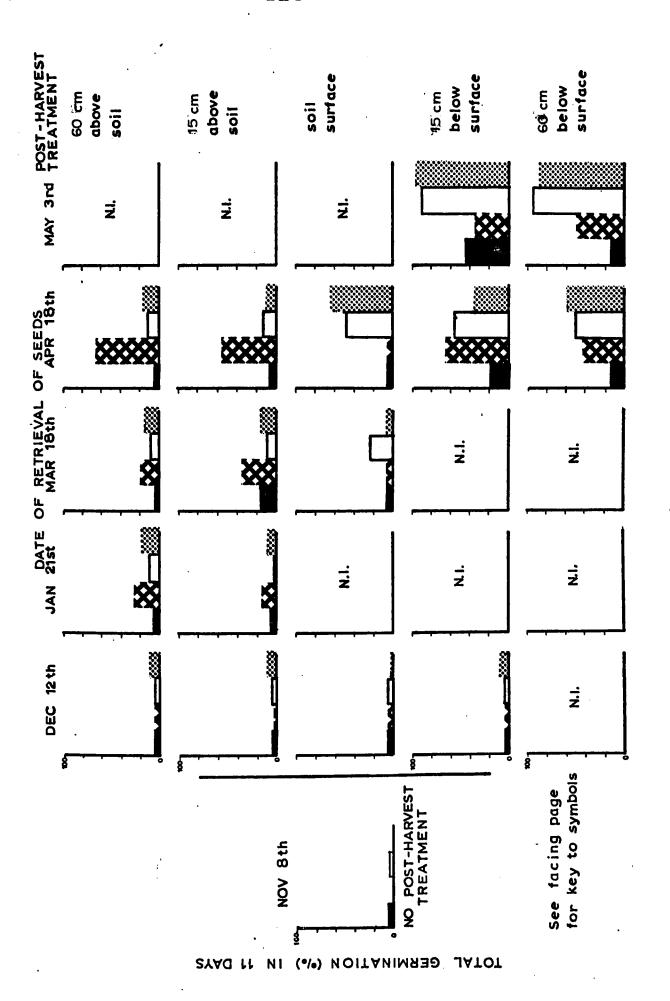
With Without With Without remains of remains of remains of remains of inflorescence inflorescence inflorescence







3 - N.I. = Treatment combination not included



of this collection that had wintered with remains of the inflorescence gave very little germination regardless of the position in which they had wintered. Seeds of collection P3 that had wintered above the soil gave a significantly lower percentage of germination than seeds that had wintered on or below the soil surface. This difference was observed among seeds that had wintered either with or without remains of the inflorescence.

## b) The influence of the inflorescence

Table A5.51 in Appendix 5 presents the comparison between germination of seeds that had wintered with and without remains of the inflorescence. In several treatments seeds that had wintered in the absence of plant remains gave higher germination than seeds that had wintered in the presence of plant remains. With collection P3 this effect was noticed in early retrievals whereas in collection P2 the effect occurred in the later retrievals.

## c) <u>Differences between collections</u>

The differences in germination response between identical treatments of collections P2 and P3 are presented in table A5.51 in Appendix 5. In general, seeds of collection P2 gave higher germination from above ground treatments and those of collection P3 gave higher germination from treatments that had wintered on or below the soil. The expression of these effects was influenced particularly by the presence or absence of inflorescence remains during overwintering.

## d) Differences between retrievals

The change in germination response with the duration of wintering is illustrated in figure 7.47. This effect was not analysed statistically since the design was incomplete and the conditions of the germination trials may have been different during the different trials. The changes in germination response that were observed were gradual increases in germination with time. These changes were dependent, to a large degree, on the particular treatment to which the seed has been subjected.

7.7.4 Discussion of the results of experiment 13

## a) <u>Experimental error</u>

The values for the error mean squares in most of the analyses of this data were larger than had been encountered This situation reflected greater in previous experiments. variation between replicates of the same treatment, and probably was a consequence of the relationship between replicates in the post-harvest treatments. experiment the seeds assigned to each replicate in the germination experiment had come from a separate replicate Thus the differences of each overwintering treatment. between these replicates, expressed in the form of experimental error, included differences in the field environment and in the environment of the germination experiment. If the seeds of each post-harvest treatment had been pooled and then divided among replicates for the germination experiment, the mean germination percentages for treatments would have been the same but the agreement between replicates would doubtless have been greater. Since seeds from post-harvest replicates were pooled in experiment 9, the greater agreement between replicates observed in that experiment probably masked the true nature of variation between replicates in post-harvest conditions. However most of the environmental and genetic influences observed in experiment 9 were confirmed in the different combinations of treatments in that experiment and thus the only major effect of masking part of the experimental variation was to increase the sensitivity of statistical analyses. This probably accounts for the smaller number of significant differences declared in this analysis than in experiment 9.

# b) <u>Differences between seeds wintering with and without</u> plant remains.

The presence or absence of remains of the parent plant with the seeds during overwintering hardly influenced the germination of seeds of collection P3 but influenced strongly the germination of seeds of collection P2.

Regardless of the position in which the seeds had remained during the winter, germination was greater in those samples which had not been in the presence of remains of the inflorescence. This difference could be observed among seeds retrieved in January and at each subsequent retrieval. The difference was more pronounced at later retrievals.

It appears that seeds of collection P2 were inhibited in their after-ripening in the presence of the remains of the

parent inflorescence. Since this effect occurred regardless of winter position it was probably a direct influence of substances produced by degradation of the remains of the inflorescence. The difference in sensitivity exhibited by seeds of the two collections may reflect a greater general sensitivity among seeds of collection P2. In support of this view, it was suggested in experiments 6 and 10 that seeds of this collection had acquired a secondary dormancy under specific conditions which had no apparent effect on the seeds of other collections.

# c) <u>Differences between seeds from different overwintering</u> positions

Seeds that were suspended 15 cm above the soil and seeds suspended 60 cm above the soil gave similar germination responses, other factors being equal. Similarly, seeds that were buried at 15 cm and seeds buried at 60 cm gave similar germination responses.

The percentage of germination of seeds of collection
P2 that had wintered above the soil was almost identical to
the percentage germination of seeds of this collection that
had overwintered beneath the soil. Seeds of this collection
that had wintered either above or below the soil in the
basence of remains of the inflorescence gave much higher
percentages of germination than similar seeds that had
wintered on the soil surface. Seeds that had wintered in
the presence of remains of the inflorescence were almost
equally dormant regardless of their position.

The percentage germination of seeds of collection P3

was almost the same for seeds that had wintered below the soil and seeds that had wintered on the soil surface. Seeds from both of these positions gave greater percentages of germination than seeds that had wintered above the soil surface.

Most of these comparisons between seeds that wintered above, below and on the soil surface could be made only between treatments retrieved on 18th April, 1969. However, there appeared to be a continuity in the responses of seeds of various treatments from consecutive retrievals and this will be discussed in the next section.

# d) <u>Differences between seeds retrieved from post-harvest</u> treatments at different times

The results of earlier experiments suggested that seeds of collection P2 were slower to after-ripen than seeds of other collections of A. powellii, in particular collection The results of this experiment confirm the opinion P3. and shed further light on the differences between the two Seeds of collection P3 began to after-ripen collections. at the same time or perhaps a little later than seeds of The rate at which after-ripening proceeded collection P2. in seeds of collection P2 was fairly constant with slightly more seeds germinating at each subsequent retrieval (ignoring those treatments in which the seeds remained A small number of seeds of collection strongly dormant). P3 had completed their after-ripening by the January retrieval, but there was no significant increase in the number until the April retrieval and only then among seeds in

certain positions. However, after-ripening of the seeds of collection P3 continued rapidly after this retrieval and was nearly complete in those treatments retrieved in May.

It is probably advantageous to the species if seeds do not complete their after-ripening until the beginning of May or later. Environmental conditions during April may occasionally be such that they would permit the germination of fully after-ripened seeds. However, it is very likely that such conditions occurring at that time would be followed by conditions that would be unfavourable for seedling survival. Thus the fewer seeds that have completed their after-ripening in April, the less likely will premature germination lead to high seedling mortality.

If seeds are completing their after-ripening at about the time when the first conditions occur that will lead to successful seedling establishment, then any environmental factors that influence the rate of after-ripening may indeed play an important role in controlling the germination of seeds. This might not be the case if the average seed had completed its after-ripening two months before the occurrence of the first conditions favourable for germination.

#### CHAPTER 8

#### CONCLUSIONS

#### 8.1 Statement

The results of this investigation contribute to two different areas of knowledge: (a) knowledge of the environmental control of seed germination in weedy species; and (b) knowledge of the taxonomy and ecology of the species that were studied.

#### 8.2 Seed germination in weedy species

#### 8.2.1 Dormancy

Seeds of each of the species were found to be dormant when freshly harvested. The nature of this dormancy was such that the minimum temperatures at which seeds would germinate were higher for freshly harvested seeds than for seeds that had wintered in the field. The nature of the temperature differences varied between the species and the details are discussed in section 8.3.3. The adaptive significance of this type of dormancy can be appreciated when the phenology of each species is related to climatic conditions.

In the vicinity of London, Ontario, the plants of these species that flower the earliest have produced their first

mature seeds at the latest by the middle of August. Most plants carry mature seeds by September. The seeds may be shed from the plant as soon as they are ripe or they may remain upon the inflorescence for sometime. After the winter the first seedlings of these species to emerge do so during May. The emergence of further seedlings occurs throughout the summer whenever soil conditions are favourable.

In this study, few of the plants that arose from seeds sown in late August were able to produce mature seeds before they were killed by frost. Thus if seeds shed in August or September germinate immediately there is little chance that the resulting plants will set seeds themselves. It can be seen from the following statistics that the mean values of mean daily temperature and total rainfall at London are higher in August and September than in May:

Month	Mean daily temperature	Mean total rainfall
May August September	12.5°C 20.2°C 15.8°C	2.72 inches 2.82 inches 3.35 inches

Seeds of several species, including Amaranthus spp., have been reported to germinate within a range of relatively high temperatures (Vegis, 1963, 1964). Without a dormancy mechanism, seeds that were able to germinate in the lower temperatures of May would also germinate in September. In Southern Ontario, climatic conditions that are unfavourable for the survival of plants of Amaranthus frequently occur soon after the end of September. These conditions would provide the selection pressure that would favour the evolution

of a dormancy mechanism of the nature described.

behaviour of freshly harvested seeds and seeds that had wintered in the field were observed within the temperature range 25°C to 35°C. These temperatures are in a higher range than the differences between mean daily air temperatures in May and September. However, seeds may be influenced by the higher than mean temperatures that occur during daylight hours and they are probably influenced more by soil temperatures than by air temperatures. The temperature of the soil is related to the air temperature but is often considerably different from it (Daubenmire, 1947). An insolated soil surface is characteristically at a higher temperature than the air above it.

The ability of freshly harvested seeds to germinate at high temperatures (35°C) may be of value in more temperate habitats or during years in which the very earliest seeds to germinate produce plants that are able to set seeds during July. In these situations it may be possible for a second generation of plants to grow and successfully produce seeds during the same season.

# 8.2.2 Factors that influence the loss of dormancy 8.2.2.1 General

The germination behaviour of seeds that had wintered in the field was not uniform but showed differences that were associated with genotype, pre-conditioning and the

nature and duration of post-harvest treatments. Also, environmental conditions at the time of germination trials were found to influence the germination behaviour of seeds. As a consequence, seeds of these species did not all germinate at the same time in the field. There are several ways in which a continuous or intermittent pattern of germination may be of value to the species:

- seeds to germinate are followed in some years by adverse conditions (particularly frost) that are lethal to the seedlings. If all of the seeds were to germinate in response to the earliest favourable conditions in some years the species might suffer local extinction. On the other hand, it may be advantageous to the species that some seeds should germinate at this time. In years in which subsequent adverse conditions do not occur, the plants that develop from these seeds will experience the longest growing season and consequently may produce the greatest number of seeds. Such plants may also compete more effectively with other weeds or with crop plants.
- 2) As weeds, these species become the object of cultivation practices. If all of the seeds were to germinate at one time there is a possibility that repeated cultivations or applications of herbicide would greatly reduce or even eliminate the population of seedlings. On the other hand, since cultivation practices rarely eliminate every single weed plant, it is advantageous that some seeds should germinate before cultivation has begun. The few plants from

such seeds that survive cultivation will be at a greater advantage in competition with the crop than plants that germinate later.

3) In some years and some situations environmental conditions may prevent any plant of a species from setting seeds. In these circumstances a reservoir of ungerminated seeds in the soil would ensure that some seeds were available to exploit the habitat in the following year.

Factors that influence the germination behaviour of seeds may contribute their effect at some time before the seeds have matured upon the parent plant or at any time until the seeds germinate in the field. Therefore it is logical to examine in a chronological order the relationships between environmental variables and seed germination.

8.2.2.2 Factor that influence the development of seeds
Another consequence of cultivation practices is that
they may stimulate the germination of further weed seeds.
Thus, in a crop that has received several cultivations there
are likely to be plants of the same species that are of a
variety of different ages.

It was shown in this investigation that seeds collected at the same time from plants of different ages differed in their ultimate germination behaviour after these seeds had wintered in the field. Seeds from younger plants were found to germinate more rapidly and at lower temperatures than seeds from older plants. The expression of this difference depended upon (a) the time at which the seeds were harvested,

(b) the nature of the post-harvest conditions, and (c) the genetic identity of the seeds. After 17 months in post-harvest conditions, seeds from plants of different ages showed no differences in germination behaviour. Thus it appeared that the age of the plant influenced the rate of after-ripening of the seeds it produced, rather than the ultimate germination behaviour of fully after-ripened seeds.

Two hypotheses can be suggested to explain these observations. Plants of different ages may possess different internal hormone balances associated with the onset of senescence (Varner, 1965) and the hormone balances may in turn influence the onset of dormancy in developing seeds (Amen, 1968). An alternative explanation can be based upon internal competition for resources. The number of seeds at any stage of development was less on younger plants than on older plants. Thus the seeds from younger plants probably experienced less competition with other seeds for nutrient materials during their development than did seeds of older plants. This difference may have influenced the development of the seeds and resulted in seeds from older plants requiring longer periods to after-ripen.

This latter hypothesis is not supported by the results obtained by Maun and Cavers (personal communication) with <a href="Rumex crispus">Rumex crispus</a>. They found that after a proportion of flowers had been removed from a plant, the seeds that were produced were larger and more frequently dormant that the seeds produced by non-thinned plants. They found also that after a proportion of the leaves had been removed from other

plants, the seeds that these plants produced were smaller and had less precise requirements for germination than seeds from non-defoliated plants. Both of these results suggest that fewer seeds of <u>Rumex crispus</u> are dormant when competition within the plant is more intense.

# 8.3.3 Factors that influence seeds between maturity and germination

Seeds may occupy different environments during the winter as a result of differences (a) in agricultural practice, and (b) in the retention of seeds on the parent plant. In Southwestern Ontario there are almost as many farmers who plough between successive crops in the autumn as there arefarmers who plough in the spring (see table 4.7). When the ground is ploughed in the autumn a large number of seeds are likely to pass the winter beneath the soil surface. Some of these seeds will have fallen from their parent plants before or during ploughing and will pass the winter without contact with plant remains. Other seeds will remain upon the inflorescences as these are ploughed into the ground. When ploughing is delayed until the spring, the majority of seeds that are shed from the plants probably pass the winter on the soil surface or are shallowly buried. Seeds that remain attached to the inflorescence will remain at different heights above the soil surface (depending upon their positions on the plant) until the plant, or part of it, falls to the ground.

In the present investigation, seeds were set to overwinter in situations intended to simulate three of the natural situations in which seeds overwinter. Striking differences were seen in the subsequent germination behaviour of these seeds. Seeds that had remained on plant remains above the soil surface were slower to after-ripen (i.e. they germinated more slowly or required higher temperatures to germinate) than seeds that had wintered away from plant remains either on or below the soil surface.

This phenomenom was investigated in greater detail with seeds of two collections of <u>A. powellii</u>. The results for one collection indicated that the difference in rates of after-ripening was associated with the presence or absence of remains of the parent plant, irrespective of the position of the seeds relative to the soil surface. The results for the other collection indicated that the difference in the rates of after-ripening was associated with contact with the soil, irrespective of the presence or absence of remains of the parent plant. Seeds were more frequently dormant after wintering in contact with remains of the parent plant and without contact with the soil in the respective collections (collections P2 and P3).

It has already been suggested that the influence of plant remains may involve the release of substances during decomposition. It is possible that such substances directly influence the seeds, perhaps by interfering with processes that are involved in the after-ripening of the seeds. It is also possible that degradation products influence after-ripening indirectly by inhibiting the activities of micro-

organisms that may normally contribute to after-ripening.

Crocker (1916) suggested that bacteria and fungi may reduce
the dormancy of seeds by enzymatic hydrolysis or decomposition
of the seed coats. He also stated that any treatment that
weakens the seed coats of A. retroflexus will break dormancy.

(This phenomenom has been demonstrated in the use of
scarification to determine the viability of ungerminated
seeds.) Rice (1964) has shown that extracts of the inflorescence
of A. retroflexus inhibit the growth of several different
species of bacteria.

The influence of contact with the soil on subsequent germination behaviour also may indicate the role of soil microorganisms. Although microorganisms are doubtless present upon seeds that remain above the ground on plant remains, they probably occur in greater numbers of individuals and species in the soil. Moreover, the environment of the soil is probably more frequently favourable for their growth and activity than is the environment of the plant inflorescence.

Crocker (1916) and Barton (1945) have stated that dry-stored seeds of A. retroflexus lose their dormancy within two or three months, and Crocker suggested that this phenomenom was related to changes in the seed coat. In the present investigation, seeds that were dry-stored for six to eight months were less frequently dormant than freshly harvested seeds but were more frequently dormant than seeds that had passed the same period of time on or below the soil surface.

Differences were found in the germination behaviour of

of seeds harvested at different times, wintered in the field, and set to germinate at the same time in the spring. Fewer seeds from an earlier harvest were dormant at alternating temperatures of 25°/10°C than seeds from a later harvest. The possibility that this difference was related to differences in preconditioning was considered unlikely since seeds that were dry-stored for identical periods did not differ in their germination behaviour. Thus it appeared that germination behaviour was influenced primarily by the nature and length of the period during which seeds were exposed to post-harvest conditions. The mechanism of this effect might be related to microbial activity. Seeds from the earlier harvest were subjected to a longer exposure to soil microorganisms and in addition, the conditions following the early harvest (August) probably were more favourable for microbial activity than the conditions following the later harvest (October).

Factors that influenced seeds between maturity and the time of germination appeared to affect the rate of after-ripening but not the ultimate response of fully after-ripened seeds. Seeds that were collected at the two different harvest times already mentioned and left in the field for 17 months showed no differences in germination behaviour that were related to time of harvest.

8.2.2.4 Factors that influence germination at the time that seeds are set to germinate

As a result of the influence of inherent and environmental factors during the development and wintering of seeds, there

will be seeds at many different stages of after-ripening when the first conditions occur in the spring that are condusive to the germination of any seeds. This conclusion can be drawn from the studies of seeds set to germinate in uniform environments. However, in one field individual seeds will occupy a variety of environments differing in characteristics of the soil, in the presence of other vegetation, and in many other ways. In addition, it is probable that fully after-ripened seeds will vary in their germination response as a result of small genetic differences between them. Consequently, there may be differences in the time of germination in the field even among seeds that have experienced identical conditions during their development and wintering. Temperatures, light conditions and soil types have been shown to influence the germination response of seeds from identical pre-treatments.

Most seeds that were dormant at low temperatures could be induced to germinate by raising the temperature. However, the temperature at which the most seeds can germinate or at which germination is the most rapid may not be the "optimum temperature" in terms of the survival of the species. Such a temperature would probably not be optimum for a species that depended for its survival upon intermittent or continuous patterns of germination.

At low temperatures, seeds of each species that had wintered in the field gave greater percentages of germination in alternating light and darkness than in continous darkness. This differences was less at higher temperatures and in

most cases did not exist at the highest temperatures that were used (40°C by day and 25°C by night). This response may be of importance in restricting germination to seeds that are on the soil surface (in light) or are buried at shallow depths (in darkness but subjected to high temperatures). The advantage of such a response might be to restrict germination to seeds that are in positions that permit successful seedling emergence and establishment. Thus, seedlings that would otherwise emerge from seeds buried at great depths (darkness, low temperatures) would probably experience high mortality rates.

The germination behaviour of seeds of each species on different soils suggested that the heat-absorbing properties of the soil are critical factors in the environment of the seed.

- 8.3 Taxonomy and ecology of the three species
- 8.3.1 Taxonomic description of the species

Until this investigation was undertaken, taxonomic descriptions of these species were almost exclusively in qualitative terms. Furthermore, most descriptions were only of use in identification when specimens of each species were on hand and character states could be compared between them. This situation is illustrated in table 3.3. In the present investigation, samples of each species from Southern Ontario were described in the quantitative terms of a number of characters. Table 3.25 on page 100 summarises the descriptions of the three species in terms of 20 of these

characters. The importance of the 20 characters in describing variation between the species was demonstrated by numerical techniques in Chapter 3.

In addition to providing a quantitative description of the species, the taxonomic study revealed patterns of variation within the species. In one species, A. powellii, a discontinuous pattern of variation was confirmed by numerical methods of analysis. This pattern was subsequently of value in explaining variation in the germination behaviour of seeds of different collections of this species. These relationships are discussed in section 8.3.4.

## 8.3.2 Geographical distribution of the species

The study of geographical distribution revealed differences in the frequency of occurrence of the species and probable differences in their habitat requirements. These differences suggested relationships between germination behaviour of the seeds, climatic conditions and the relative success of each species in Southern Ontario. The relationships are discussed in the following sections.

## 8.3.3 Germination behaviour of the seeds

The relationships between environmental and inherent variables and germination behaviour (discussed in section 8.2) were basically the same for seeds of each species.

Differences that were observed between species were mainly differences of degree rather than differences in the nature

of the relationships.

Freshly harvested seeds of A. hybridus and A. retroflexus germinated more rapidly and at lower temperatures (30/15°C) than seeds of most collections of A. powellii (which with the exception of one collection did not germinate below 35/20°C). In field trials only a very small number of freshly harvested seeds germinated and these were nearly all of A. retroflexus. Thus there appeared to be little significance in the differences in germination behaviour of the freshly harvested seeds of different species.

After wintering under field conditions, seeds of

A. powellii germinated generally before (or at lower
temperatures than) seeds of A. retroflexus while seeds of

A. hybridus were often the last to germinate. The differences
in germination between seeds of A. hybridus and those of the
other species were observed also in field trials and in most
cases the differences were more pronounced under these
conditions. The relationship between germination of the seeds of

A. powellii and that of the seeds of A. retroflexus showed
considerable variation among the different experiments.

The differences between species in the temperature requirements for germination of their seeds appear to reflect differences in the widespread distributions of the species.

Amaranthus hybridus has a distribution than includes much warmer regions than are included in the distributions of either A. powellii or A. retroflexus and seeds of A. hybridus require higher temperatures for germination than seeds of the other species.

- 8.3.4 Characteristics of each species
- 8.3.4.1 Amaranthus hybridus

Possible explanations for the discontinuous distribution and low frequency of this species were given in Chapter 4. The results of the germination studies provided further insights into the status of this species. It was seen that seeds of this species were slowest to germinate, particularly in field trials. Also, there was evidence that these seeds required higher temperatures for their germination than seeds of the other two species. As a result, plants of this species would begin their growth later in the season than plants of the other two species. When plants of the three species were grown from seeds that had germinated at the same time, it was seen that plants of A. hybridus took longer to flower and produce ripe seeds than plants of the other species. These two observations probably explain why the difference in temperature requirements for germination between freshly-harvested and wintered seeds of this species was less than that of seeds of A. powellii: (1) Seeds of A. hybridus require high temperatures for germination; (2) Seeds are produced late in the season when such temperatures are unlikely to occur.

The evidence suggests that the length of the growing season may be a more important limiting factor for this species in this region than for the other two species. It was suggested in Chapter 4 that the association of this species with sandy soils might be related to differences in the growing season available to plants growing on different soils. The germination behaviour of seeds of this species on

different soils supports this view. Germination of seeds occurred earlier and more frequently on Brokkston sandy. loam than on any of the other soils included. It appeared that the properties of this soil would enable it to warm up in the spring more rapidly than the other soils.

#### 8.3.4.2 Amaranthus powellii

Characteristic of this species was the ability of its seeds to germinate more rapidly at low temperatures after wintering than seeds of the other species. This was in contrast to the germination behaviour of freshly harvested seeds of this species, which required higher temperatures for germination than seeds of the other species.

The existence of a pronounced dormancy mechanism among the seeds of this species can be compared with the explanation offered for the less pronounced dormancy of seeds of the previous species. Plants of this species mature more rapidly than plants of A. hybridus and seeds of this species germinate earlier in the season. As a result plants of this species set seeds earlier in the growing season than plants of A. hybridus, and at a time when temperatures are high enough to permit the germination of non-dormant seeds. A dormancy mechanism is necessary to prevent germination at this time.

In germination trials in the field, seeds of A. powellii germinated more rapidly and in greater numbers than seeds of A. retroflexus. However, in the study of distribution, the

frequency of occurrence of <u>A. powellii</u> was found to be less than that of <u>A. retroflexus</u>. If these two observations are related, they may illustrate a point raised earlier: An optimum germination response, from the ecological viewpoint, may not involve the most rapid germination or the greatest percentage of germination in any particular situation. The value to a weedy species of a continuous or intermittent pattern of germination has already been discussed. Thus, it appears that <u>A. retroflexus</u> shows greater adaptation to the weed habit in its germination response than does <u>A. powellii</u>. Perhaps this difference is explained by the reports (e.g. Sauer, 1967) that <u>A. powellii</u> has become a widespread weed only recently.

Germination behaviour was studied with seeds of six different collections of this species. Three different patterns of response were discerned and these corresponded to taxonomic relationships observed within the species.

Seeds of collections of 'type B' and of most collections of 'type A' were dormant at temperatures as high as 35°/20°C when freshly harvested. Seeds of 'type B' after-ripened more rapidly than seeds of these same collections of 'type' A' but fully after-ripened seeds of the latter type germinated more rapidly than similar seeds of the former type. Seeds of collection Pl differed in their responses from seeds of each of the above mentioned collections.

In particular, freshly harvested seeds of this collection germinated at temperatures as low as 30°/15°C. Plants of this collection were initially considered to be of 'type A'

but in Chapter 3 it was pointed out that when plants were grown under experimental field conditions, those of collection Pl possessed a combination of characters that differed from those possessed by plants of collections of 'type A' and 'type B'.

The significance of these differences cannot be determined from this investigation since little is known of the origins, distributions or frequencies of the variants. However, knowledge of this variation provides a basis from which further work can be developed.

## 8.3.4.3 Amaranthus retroflexus

The time at which seeds of this species germinated in field trials and the rate of subsequent development of the plants (both were almost the same as those of A. powellii) suggested that seeds of this species might possess a dormancy mechanism similar to that of seeds of A. powellii (in order to prevent the germination of freshly-matured seeds in late summer). The results of these investigations indicated that seeds of this species possess a slightly different type of dormancy from that of seeds of A. powellii. Freshly-harvested seeds of this species germinated at alternating temperatures as low as 250/100c. However, the rate of germination of freshly harvested seeds at these temperatures was much lower than the rate of germination at the same temperatures of seeds that had wintered in the field. Thus, although temperatures in late summer may be high enough to permit germination of seeds, it is probable that they rarely remain at this level long enough for germination to occur.

The statement often has been made that seeds of Amaranthus retroflexus are inhibited in their germination by light (Crocker, 1916, Evans, 1922, Barton, 1945, and Rojas-Garciduenas and Kommedahl, 1960). In most of these reports, the seeds referred to were freshly-harvested or had been dry-stored. In the present investigation seeds of two collections of A. retroflexus that had been dry-stored gave a greater percentage of germination in alternating light and darkness than in continuous darkness. Seeds that remained dormant in continuous darkness germinated when they were transferred to alternating light and darkness. Similar responses were noted for seeds that had wintered in the field. The apparent disagreement with earlier reports probably reflects a difference between the effects of continuous light and alternating light and darkness. The significance of such a difference has not been reported.

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#### 8.4 Relevance of the results

It can be asked of any experimental work whether the results are relevant to the non-experimental situation. In the experiments reported here several precautions were taken to ensure that the results would be as meaningful as possible. Germination trials in which freshly-harvested seeds were used were included in order to determine the probability that seeds will germinate in late summer, when first produced. In all other experiments the germination behaviour of seeds was examined only after the seeds had been subjected to a post-harvest treatment. With the exception of

experiment 13, all of the germination trials were conducted at times when germination could be expected to occur naturally. The seeds used in each experiment (except in preliminary experiments) were taken from plants that were grown under uniform field conditions at the Department of Botany Experimental Farm. Pre-harvest and post-harvest treatments were planned to simulate the different conditions that might be experienced by seeds in the field rather than to study the effects of individual variables. Once it has been established that conditions actually encountered by seeds influence subsequent behaviour then there is justification for studying the effects of isolated variables.

There are several ways in which the conditions employed in these experiments departed from natural conditions and it is worthwhile to consider how these departures may have influenced the results. Although seeds were taken from plants grown under field conditions, the nature of these conditions was different from those that might be experienced by an agricultural weed. The plants were grown in rows and were spaced about a metre apart and consequently they were free from competition with other plants from the time they were planted until maturity. It is possible that responses of seeds to climatic factors (which might ultimately influence the seed) under these conditions might be completely masked under natural conditions by greater responses to competition for light, water and nutrients. This possibility has been discussed by Evans (1963).

The greatest departures from natural conditions accompanied the ultimate germination trials. In planning these trials two alternative approaches were conceived.

One approach was to attempt to reproduce natural conditions by conducting germination trials in the field. The other approach was to use arbitrary, but defined conditions in the laboratory or greenhouse.

At first field trials seem the more appropriate.

However, fields vary enormously, one from another, and field conditions change continually during the experiment and differ from year to year. To be truly representative, it would be necessary to repeat identical experiments under an array of different field conditions during several successive years. Other problems arise with field trials from the need to distinguish the seeds under observation from seeds of the same species that occur naturally at the experimental site. One attempt to solve this problem involves sowing seeds into plots of sterilised soil. However, sterilisation may alter subtly the characteristics of the soil. Moreover, after the seeds have been sown into the plots some may be washed or blown out of the plots while foreign seeds may be washed or blown in.

The use of controlled conditions in the laboratory has much to offer in comparison with field conditions. It is relatively easy to define conditions with precision and to repeat them. The heterogeneity of the experimental environment is less than in the field, allowing differences between experimental treatments to be detected with greater sensitivity. It is easier in the laboratory to control

different environmental variables independently, thereby permitting greater insight into the mechanisms that control germination. A further advantage is that conditions known to be optimum for germination can be ensured from the moment the experiment is begun.

The most obvious disadvantage to the use of controlled conditions lies in the choice of conditions. Even when this choice is made with great care there is the risk that the set of conditions chosen are such that they would never be encountered by seeds in the field. For example, it is tempting to maintain a defined set of conditions until germination is complete and this may be a matter of weeks. However, in natural situations conditions frequently are changing from one day to the next and during a diurnal cycle. It is possible that one of the important factors controlling germination is the day to day variability of the environment.

Since both field and laboratory conditions are accompanied by advantages and disadvantges of different forms, the most compelling solution to this problem is to use both approaches. This solution was employed in this investigation with interesting results. Comparisons of the results of germination trials in the two types of conditions allowed the following generalisations to be made: (1) laboratory trials were more sensitive for determining differences in germination behaviour; and (2) for the most part, differences observed in laboratory trials were

confirmed in field trials, although they were often of a different order of magnitude in the two trials. The latter conclusion is important since it allows a greater confidence to be placed in the results of all of the germination trials that were conducted in the laboratory.

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# APPENDIX 1

# TAXIMETRIC DATA

The data are arranged by samples (see Table 3.2) and within each sample values are presented in order for each of the 41 characters described in table 3.4.

# Sample 1:

1.04	0.86	1.27	1.13	0.97 0.21 32.25 7.00	0.13 19.36	0.01 0.67	0.00 0.82	12.00 31.00	1.64 0.00
	8.00	8.00	7.00	7.00	6.00	10.00	6.00.	0.96	1.20
54.00									

# Sample 2:

10.75 10.00 3.13 1.08 0.90 1.36 0.00 1.27 0.02 6.00 9.00 8.00	0.29	0.13 28.50	0.00	0.02	0.00 0.82	7.07	0.00
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# Sample 3:

10.88 10.00 1.11 0.88 0.00 0.55 7.00 9.00 30.00	2.41 3 0.09 (	1.43	0.21	0.13 38.41	0.00 0.63	0.00	15.98 17.00	0.00
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# Sample 4:

1.16 0.00	0.91 1.55	0.27	0.86	0.00 29.75	0.00 9.36	23.00 0.04 0.65 6.00	0.00	9.00	0.41
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# Sample 5:

1.08	0.89 0.155	0.50	1.55	0.80 0.13 38.63 16.00	0.00	0.02 0.54	0.00	5.70 36.00	0.50	
37.00										

# Sample 6:

1.08 0.00 6.00	0.94	2.50 0.02	0.29	0.76 0.13 32.38 19.00	0.13 3.13	0.07 0.61	0.00	2.79 103.0	1.55 0.00
25.00									

# Sample 7:

# Sample 8:

0.93	0.95 0.70 8.00	0.41	0.84	0.41 25.13	0.00 3.84	19.38 0.01 0.74 33.00	<b>0.00</b> 0.77	1.13 52.00	0.00
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# Sample 9:

10.25 11.13 3 0.92 0.98 0 0.00 0.98 0 10.00 11.00 4 14.00	0.50 0.70 0.00 0.00	0.13 25.75	0.13 18.21	0.00	0.00	78.00	0.96
-----------------------------------------------------------------------	------------------------	---------------	---------------	------	------	-------	------

# Sample 10:

0.94	0.92	0.84	0.84	1.00 0.21 34.38 18.00	0.13	0.64	0.84	41.00	0.00
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# Sample 11:

10.00 11.13 2.75 5.25 0.91 0.95 0.29 1.55 0.00 0.79 0.00 0.00 7.00 12.00 27.00 9.00	0.50 0.50	0.64	0.89	102.0	0.00
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#### Sample 12:

9.63 10.38 2.88 5.13 0.93 0.00 26.75 32.25 0.50 30.63 0.93 0.95 0.55 0.27 0.13 0.13 0.01 0.00 9.07 0.79 0.00 1.41 0.01 0.00 36.63 34.55 0.55 0.73 117.0 0.00 7.00 11.00 23.00 8.00 24.00 7.00 33.00 8.00 0.86 1.24 14.00

#### Sample 13:

1.00 0.00 21.75 32.00 0.53 0.21 0.57 0.00 0.00 6.50 30.00 4.50 8.63 9.75 **2.7**5 0.86 0.79 1.13 0.89 0.94 51.00 0.12 0.29 0.02 0.00 30.00 20.57 0.61 0.72 0.00 13.00 5.00 10.00 5.00 1.03 0.86 11.00 12.00 6.00 7.00 21.00

#### Sample 14:

9.00 10.00 2.75 5.00 1.00 0.00 20.63 33.00 0.53 30.63 0.90 0.93 0.86 0.29 0.21 0.00 0.00 0.00 3.13 0.57 0.00 0.55 0.01 0.00 29.25 37.93 0.65 0.71 67.00 0.40 6.00 10.00 13.00 4.00 14.00 6.00 16.00 6.00 0.91 1.12 24.00

#### Sample 15:

12.63 10.03 2.88 5.00 0.87 0.80 43.50 33.25 0.51 30.00 1.25 0.90 1.13 0.70 0.13 0.00 0.05 0.05 33.14 2.21 0.00 2.00 0.02 0.00 51.88 96.13 0.44 0.84 105.0 0.00 12.00 19.00 38.00 16.00 38.00 15.00 38.00 14.00 1.00 1.00 13.00

#### Sample 16:

12.63 10.38 3.13 5.00 0.90 0.75 43.13 33.88 0.54 30.00 1.22 0.89 0.55 0.55 0.13 0.00 0.02 0.02 45.84 0.98 0.00 0.57 0.01 0.00 49.50 115.71 0.46 0.87 94.00 0.00 13.00 17.00 25.00 11.00 35.00 15.00 43.00 16.00 0.73 1.25 11.00

#### Sample 17:

12.88 10.50 3.13 5.00 0.92 0.72 45.50 33.88 0.53 29.75 1.23 0.88 0.98 0.57 0.13 0.00 0.01 0.01 8.57 0.98 0.00 0.79 0.02 0.00 55.00 118.86 0.42 0.83 59.00 0.24 17.00 18.00 30.00 15.00 32.00 14.00 34.00 17.00 0.94 1.06 14.00

#### Sample 18:

14.00 10.38 3.25 5.00 0.85 0.77 39.25 36.68 0.57 31.25 1.35 0.86 0.86 0.55 0.50 0.00 0.04 0.02 26.21 0.84 0.00 0.79 0.01 0.00 57.50 64.29 0.42 0.68 42.00 0.00 14.00 19.00 26.00 18.00 23.00 17.00 27.00 16.00 1.03 1.07 14.00

# Sample 19:

13.38 10.63 3.13 5.00 0.92 0.60 39.88 34.50 0.55 29.00 1.27 0.84 1.41 1.13 0.13 0.00 0.02 0.05 22.70 3.43 0.00 1.43 0.04 0.00 45.25 99.07 0.53 0.88 56.00 0.00 13.00 16.00 26.00 17.00 26.00 12.00 32.00 13.00 0.93 1.14 18.00

#### Sample 20:

14.13 9.63 3.00 5.00 0.75 0.87 41.25 35.00 0.56 30.50 1.50 0.87 0.41 1.98 0.00 0.00 0.04 0.03 17.64 2.00 0.00 0.29 0.06 0.00 50.25 77.93 0.47 0.82 136.0 0.18 13.00 22.00 59.00 18.00 48.00 15.00 88.00 18.00 0.91 1.35 12.00

## Sample 21:

13.88 10.75 3.00 5.00 0.70 0.90 39.50 34.38 0.53 30.50 1.30 0.89 0.41 0.79 0.00 0.00 0.09 0.01 16.29 1.70 0.00 0.86 0.02 0.00 50.50 54.00 0.49 0.78 80.00 0.00 9.00 21.00 22.00 17.00 35.00 13.00 39.00 15.00 0.69 1.22 16.00

#### Sample 22:

14.63 10.25 3.00 4.88 0.80 0.84 39.75 35.88 0.52 31.63 1.45 0.88 0.84 1.36 0.00 0.13 0.06 0.01 13.36 2.98 0.00 2.27 0.05 0.00 52.13 56.70 0.48 0.76 102.0 0.00 17.00 20.00 28.00 10.00 60.00 16.00 46.00 16.00 0.63 1.03 11.00

#### Sample 23:

14.50 10.75 2.88 5.13 0.83 0.85 44.50 35.88 0.53 31.38 1.36 0.87 0.86 1.36 0.13 0.13 0.05 0.02 11.71 1.55 0.00 1.13 0.03 0.00 49.00 101.71 0.52 0.91 59.00 0.00 18.00 22.00 28.00 15.00 29.00 15.00 33.00 13.00 0.93 1.10 10.00

#### Sample 242

13.50 8.63 3.00 5.00 0.85 0.60 39.50 33.25 0.58 28.38 1.58 0.85 1.43 0.55 0.00 0.00 0.02 0.01 25.71 1.07 0.00 0.84 0.05 0.00 58.13 92.98 0.38 0.68 34.00 0.00 14.00 17.00 28.00 12.00 22.00 11.00 21.00 11.00 1.18 0.89 37.00

# Sample 25:

12.50 9.63 3.00 5.00 0.50 0.57 32.38 32.60 0.63 27.25 1.31 0.91 0.86 1.41 0.00 0.00 0.06 0.06 13.70 2.27 0.02 1.64 0.02 0.04 49.75 57.36 0.44 0.65 49.00 0.00 12.00 17.00 23.00 10.00 23.00 11.00 42.00 11.00 0.78 1.43 12.00

### Sample 26:

3.00 4.88 0.59 0.00 33.13 32.13 0.55 13.00 8.13 1.60 0.89 0.57 0.41 0.00 0.13 0.06 0.00 15.27 1.55 0.00 0.86 0.01 0.00 49.75 12.79 0.42 0.67 40.00 0.00 7.00 13.00 14.00 5.00 17.00 7.00 20.00 10.00 0.82 1.18 22.00

#### Sample 27:

13.88 8.50 3.00 5.00 0.47 0.00 34.50 33.38 0.57 29.00 1.64 0.87 0.41 0.57 0.00 0.00 0.03 0.00 7.71 1.70 0.00 1.71 0.01 0.00 48.25 29.36 0.46 0.72 38.00 0.00 11.00 14.00 12.00 6.00 16.00 8.00 14.00 18.00 0.86 1.00 32.00

# Sample 28:

13.25 8.88 3.00 5.00 0.95 0.70 39.88 33.63 0.56 29.88 1.52 0.89 0.21 1.84 0.00 0.00 0.01 0.01 16.70 0.84 0.00 0.70 0.05 0.00 51.50 146.0 0.43 0.77 37.00 0.00 13.00 17.00 17.00 10.00 22.00 14.00 23.00 12.00 0.82 1.11 22.00

#### Sample 29:

13.25 9.25 3.00 5.00 0.90 0.85 41.00 33.75 0.56 28.25 1.44 0.84 0.79 0.21 0.00 0.00 0.05 0.02 8.00 2.21 0.00 11.07 0.02 0.01 59.13 14.98 0.38 0.69 60.00 0.00 16.00 20.00 27.00 15.00 41.00 17.00 35.00 16.00 0.79 1.02 6.00

#### Sample 30:

14.00 10.88 2.75 5.00 0.85 0.77 42.75 36.63 0.51 30.88 1.30 0.84 0.57 1.55 0.21 0.00 0.08 0.03 17.36 0.55 0.00 13.27 0.03 0.01 53.38 49.41 0.47 0.80 47.00 0.00 15.00 18.00 24.00 12.00 28.00 13.00 35.00 16.00 0.83 1.21 19.00

# Sample 31:

14.38 9.88 3.00 5.00 0.92 0.60 48.38 32.63 0.52 28.13 1.46 0.86 0.55 0.41 0.00 0.00 0.01 0.03 19.41 1.41 0.00 0.41 0.00 0.00 51.13 159.84 0.47 0.95 13.00 0.00 15.00 15.00 12.00 7.00 17.00 11.00 15.00 11.00 0.82 1.02 28.00

# Sample 32:

13.13 9.13 2.88 5.00 0.85 0.77 36.13 32.13 0.54 28.75 1.46 0.90 0.98 1.27 0.13 0.00 0.02 0.02 12.98 1.27 0.00 9.07 0.04 0.01 46.88 137.27 0.47 0.77 33.00 0.00 13.00 17.00 17.00 14.00 21.00 10.00 21.00 15.00 0.86 1.07 24.00

### Sample 33:

13.75 9.00 3.00 5.00 0.75 0.90 36.75 34.75 0.54 30.00 1.55 0.86 0.79 2.00 0.00 0.00 0.03 0.01 28.50 3.37 0.00 0.86 0.04 0.00 46.25 99.36 0.49 0.79 19.00 0.00 13.00 13.00 13.00 8.00 24.00 12.00 25.00 16.00 0.63 1.21 31.00

### Sample 34:

14.25 9.88 3.13 4.88 1.00 0.71 42.00 36.38 0.55 31.50 1.45 0.87 1.64 0.70 0.13 0.13 0.00 0.04 12.57 0.27 0.00 0.29 0.03 0.00 61.63 58.27 0.39 0.68 53.00 0.00 17.00 20.00 26.00 14.00 25.00 16.00 22.00 16.00 1.07 0.90 15.00

#### Sample 35:

13.50 9.38 2.75 5.00 0.77 0.65 42.13 33.63 0.53 29.13 1.45 0.87 0.29 0.55 0.21 0.00 0.05 0.04 18.70 0.84 0.00 0.13 0.03 0.00 62.25 177.64 0.37 0.68 29.00 0.00 15.00 18.00 26.00 15.00 31.00 15.00 36,00 16.00 0.84 1.16 17.00

#### Sample 36:

13.63 8.75 2.88 4.88 0.70 0.81 36.63 33.88 0.52 29.00 1.57 0.86 0.55 1.07 0.13 0.41 0.10 0.12 16.27 0.98 0.00 1.43 0.03 0.00 49.25 96.50 0.45 0.74 25.00 0.00 12.00 14.00 20.00 10.00 19.00 10.00 18.00 12.00 1.05 0.95 23.00

#### Sample 37:

14.25 9.75 3.00 5.00 0.87 0.77 41.38 34.25 0.53 29.75 1.48 0.87 1.07 1.07 0.00 0.00 0.02 0.01 20.84 0.79 0.00 0.21 0.06 0.00 60.50 54.86 0.40 0.68 92.00 0.00 15.00 20.00 37.00 13.00 43.00 18.00 49.00 16.00 0.86 1.14 15.00

#### Sample 38:

15.13 10.75 3.13 4.75 0.97 0.76 42.75 36.38 0.55 30.75 1.42 0.85 0.41 0.79 0.13 0.21 0.01 0.03 21.36 1.13 0.00 0.50 0.02 0.00 53.88 15041 0.48 0.79 96.00 0.00 18.00 25.00 40.00 13.00 49.00 17.00 46.00 18.00 0.89 1.02 10.00

#### Sample 39:

13.38 9.38 3.00 5.00 0.72 0.82 38.25 25.00 0.57 30.38 1.45 0.87 1.41 1.13 0.00 0.00 0.06 0.05 8.21 1.71 0.00 0.55 0.05 0.00 50.50 24.00 0.45 0.76 49.00 0.00 12.00 17.00 17.00 12.00 22.00 13.00 28.00 13.00 0.76 1.25 15.00

#### Sample 40:

12.63 10.00 2.88 5.00 0.97 0.82 42.88 33.13 0.52 28.50 1.27 0.86 0.55 0.86 0.13 0.00 0.01 0.03 21.27 0.70 0.00 0.57 0.02 0.00 53.25 16.50 0.42 0.81 57.00 0.00 13.00 18.00 24.00 12.00 33.00 13.00 36.00 13.00 0.77 1.16 16.00

### Sample 41:

11.25 9.50 3.00 4.88 0.80 0.82 34.38 32.00 0.58 27.25 1.24 0.85 1.07 0.29 0.00 0.13 0.07 0.03 12.84 0.57 0.00 0.50 0.02 0.00 48.88 84.98 0.43 0.70 43.00 0.00 13.00 16.00 14.00 8.00 20.00 9.00 22.00 10.00 0.75 1.18 25.00

### Sample 42:

15.13 10.00 3.00 5.00 0.97 0.87 45.88 33.25 0.53 29.13 1.53 0.88 1.84 0.86 0.00 0.00 0.01 0.02 12.13 0.50 0.00 0.41 0.06 0.00 64.50 99.43 0.39 0.71 48.00 0.00 14.00 20.00 20.00 13.00 32.00 14.00 28.00 12.00 0.75 1.05 23.00

### Sample 43:

14.38 9.38 3.13 5.00 0.97 0.87 45.38 32.50 0.52 28.88 1.54 0.89 0.84 0.55 0.13 0.00 0.01 0.02 5.98 0.86 0.00 1.27 0.02 0.00 56.75 1/2.21 0.42 0.80 50.00 0.00 14.00 18.00 19.00 10.00 31.00 12.00 19.00 12.00 0.83 0.83 33.00

#### Sample 44:

14.88 9.13 3.13 5.00 0.97 0.67 48.00 32.63 0.59 28.13 1.64 0.86 0.70 0.70 0.13 0.00 0.01 0.01 9.71 2.55 0.00 1.27 0.02 0.00 62.75 129.36 0.38 0.76 66.00 0.00 13.00 18.00 21.00 13.00 27.00 14.00 22.00 13.00 0.90 0.94 24.00

## Sample 45:

14.25 9.63 3.00 5.00 0.85 0.82 36.00 34.50 0.54 29.63 1.49 0.86 0.50 0.84 0.00 0.00 0.03 0.03 11.71 3.43 0.00 1.70 0.01 0.00 45,25 123.93 0.53 0.80 51.00 0.00 12.00 16.00 25.00 12.00 25.00 14.00 27.00 14.00 0.97 1.05 16.00

# Sample 46:

13.99 10.38 3.00 5.00 0.97 0.87 43.13 34.00 0.57 29.63 1.34 0.87 1.55 0.55 0.00 0.00 0.01 0.01 20.13 0.86 0.00 0.55 0.02 0.00 58.00 52.29 0.42 0.74 41.00 0.00 17.00 20.00 24.00 12.00 26.00 12.00 29.00 16.00 0.91 1.10 14.00

# Sample 47:

13.75 9.38 3.00 5.00 0.87 0.72 39.50 32.50 0.56 28.00 1.47 0.86 1.07 0.27 0.00 0.00 0.01 0.07 5.43 2.57 0.00 2.29 0.02 0.00 51.00 9.43 0.45 0.77 39.00 0.00 16.00 21.00 28.00 13.00 33.00 18.00 44.00 15.00 0.80 1.26 8.00

# Sample 48:

13.63 9.13 3.00 5.00 0.90 0.72 34.38 37.38 0.53 32,88 1.51 0.88 1.41 0.98 0.00 0.00 0.02 0.03 9.98 1.13 0.00 1.55 0.06 0.00 41.38 73.41 0.55 0.83 24.00 0.00 15.00 15.00 24.00 11.00 28.00 13.00 31.00 10.00 0.87 1.12 11.00

#### Sample 49:

13.63 10.00 2.75 4.88 0.92 0.75 42.13 36.13 0.52 32.63 1.37 0.91 0.27 0.57 0.21 0.13 0.02 0.03 12.41 6.13 0.00 1.13 0.02 0.00 52.13 137.27 0.45 0.81 28.00 0.00 10.00 13.00 24.00 13.00 26.00 11.00 27.00 12.00 0.94 1.05 16.00

#### Sample 50:

12.38 8.25 3.13 5.00 0.82 0.92 33.63 33.25 0.53 28.50 1.51 0.86 0.84 0.21 0.13 0.00 0.03 0.01 12.84 3.36 0.00 3.14 0.03 0.00 45.00 28.00 0.46 0.75 16.00 0.00 13.00 13.00 14.00 8.00 7.00 4.00 10.00 6.00 1.35 0.97 51.00

# Sample 51:

13.50 9.38 3.13 4.75 0.76 0.87 37.00 43.63 0.53 30.00 1.45 0.87 0.57 0.55 0.13 0.21 0.02 0.01 14.57 1.13 0.00 0.86 0.02 0.00 38.75 56.50 0.59 0.95 28.00 0.00 12.00 12.00 10.00 8.00 11.00 8.00 15.00 10.00 0.83 1.25 27.00

# Sample 52:

14.50 9.00 2.88 5.00 0.90 0.80 38.88 35,88 0.56 29.88 1.67 0.83 1.71 2.57 0.13 0.00 0.01 0.02 42.13 1.55 0.00 0.98 0.18 0.00 43.50 64.00 0.54 0.89 36.00 0.00 12.00 14.00 13.00 10.00 14.00 10.00 17.00 9.00 0.89 1.16 32.00

# Sample 53:

13.88 9.00 3.13 5.00 0.90 0.80 41.63 35.75 0.53 30.63 1.55 0.86 1.84 0.86 0.13 0.00 0.02 0.02 27.41 0.50 0.00 0.84 0.05 0.00 52.50 87.71 0.44 0.79 37.00 0.00 13.00 15.00 18.00 12.00 19.00 11.00 21.00 11.00 0.93 1.09

## Sample 54:

12.38 9.38 3.00 5.00 0.80 0.80 38.00 34.13 0.54 29.50 1.34 0.86 1.41 1.13 0.00 0.00 0.02 0.02 11.43 1.27 0.00 1.14 0.04 0.00 56.63 68.55 0.38 0.67 42.00 0.00 13.00 18.00 16.00 12.00 16.00 14.00 19.00 14.00 0.94 1.12 14.00

#### Sample 55:

13.50 9.00 3.00 5.13 0.88 0.80 41.13 34.88 0.59 28.75 1.51 0.83 0.86 0.57 0.00 0.13 0.01 0.02 36.70 4.13 0.00 0.50 0.03 0.00 57.88 138.41 0.39 0.71 53.00 0.00 14.00 16.00 26.00 11.00 32.00 12.00 37.00 12.00 0.82 1.17 16.00

## Sample 56:

13.63 9.00 2.75 4.88 0.84 0.66 38.88 35.25 0.55 30.13 1.54 0.85 1.70 1.43 0.21 0.13 0.05 0.01 14.70 1.07 0.00 0.70 0.07 0.00 50.88 20.98 0.44 0.76 31.00 0.00 13.00 15.00 17.00 18.00 15.00 13.00 19.00 9.00 1.00 1.12 22.00

## Sample 57:

12.50 9.25 3.00 5.00 0.82 0.62 40.13 34.50 0.57 29.25 1.35 0.85 1.43 0.21 0.00 0.00 0.05 0.03 12.41 0.86 0.00 0.50 0.02 0.00 49.13 77.55 0.44 0.82 49.00 0.00 13.00 18.00 20.00 12.00 25.00 12.00 33.00 14.00 0.77 1.27 19.00

# Sample 58:

13.50 9.00 3.13 5.00 0.85 0.80 39.38 33.38 0.53 29.75 1.51 0.89 1.43 0.57 0.13 0.00 0.03 0.02 5.13 1.13 0.00 0.50 0.05 0.00 43.50 55.14 0.52 0.91 30.00 0.00 14.00 16.00 19.00 18.00 22.00 10.00 22.00 11.00 0.90 1.05 32.00

# Sample 59:

14.00 9.25 3.25 4.88 1.00 0.87 41.63 34.14 0.53 30.38 1.53 0.89 0.57 0.79 0.50 0.13 0.00 0.01 28.84 1.27 0.00 0.55 0.03 0.00 47.63 78.84 0.49 0.87 40.00 0.00 13.00 17.00 17.00 10.00 22.00 13.00 25.00 12.00 0.80 1.17 22.00

# Sample 61:

13.63 9.00 3.00 5.00 0.80 0.95 36.13 34.75 0.55 29.63 1.54 0.85 0.84 1.71 0.00 0.00 0.06 0.01 15.27 0.50 0.00 2.84 0.05 0.00 48.50 23.14 0.47 0.74 40.00 0.00 14.00 14.00 15.00 9.00 16.00 12.00 18.00 9.00 0.92 1.10 23.00

# Sample 62:

12.13 8.63 3.00 5.00 0.60 0.87 35.38 33.00 0.55 28.50 1.42 0.86 0.98 0.55 0.00 0.00 0.01 0.02 20.55 0.86 0.00 0.29 0.05 0.00 46.13 87.27 0.45 0.77 30.00 0.00 16.00 19.00 17.00 12.00 28.00 15.00 29.00 12.00 0.69 1.18 19.00

# Sample 63:

13.50 9.50 3.00 5.00 0.85 0.65 34.75 32.75 0.56 27.88 1.43 0.85 4.00 0.57 0.00 0.00 0.05 0.08 24.50 1.36 0.00 0.70 0.05 0.00 57.88 25.27 0.40 0.60 34.00 0.00 15.00 16.00 18.00 10.00 23.00 14.00 28.00 13.00 0.78 1.22 20.00

# Sample 64:

11.63 8.63 3.00 5.00 0.87 0.57 31.00 31.75 0.55 26.88 1.36 0.85 0.84 0.84 0.00 0.00 0.02 0.11 2.86 0.50 0.00 0.13 0.02 0.00 48.38 73.41 0.42 0.64 93.00 0.00 12.00 15.00 23.00 10.00 39.00 8.00 42.00 11.00 0.66 1.21 21.00

# Sample 65:

12.38 9.00 3.00 5.00 0.92 0.70 33.13 32.50 0.53 27.38 1.40 0.84 1.98 1.14 0.00 0.00 0.02 0.07 4.98 0.57 0.00 0.55 0.09 0.00 53.00 54.29 0.40 0.63 65.00 0.42 11.00 14.00 21.00 13.00 31.00 13.00 22.00 12.00 0.85 0.89 15.00

# Sample 66:

11.13 8.00 2.88 4.13 0.32 0.00 24.50 34.13 0.55 29.50 1.40 0.87 0.98 0.57 0.13 0.70 0.07 0.00 22.00 3.55 0.00 1.71 0.03 0.00 45.25 46.79 0.42 0.54 50.00 0.00 9.00 13.00 23.00 9.00 25.00 9.00 28.00 8.00 0.91 1.11 10.00

# Sample 67:

14.13 9.50 3.00 4.00 0.40 0.00 32.88 36.38 0.57 30.50 1.49 0.84 1.84 0.29 0.00 0.29 0.05 0.00 7.84 1.70 0.00 2.00 0.02 0.00 47.25 57.36 0.50 0.70 46.00 0.00 10.00 13.00 17.00 9.00 19.00 10.00 12.00 10.00 1.06 0.75 17.00

# Sample 68:

14.88 10.00 2.88 4.13 0.42 0.00 35.75 37.88 0.60 29.50 1.49 0.78 0.70 0.29 0.13 0.70 0.02 0.00 20.50 0.98 0.00 0.86 0.01 0.00 57.88 122.70 0.43 0.62 68.00 0.21 11.00 15.00 18.00 12.00 29.00 14.00 35.00 13.00 0.66 1.28 5.00

# Sample 69:

14.88 9.38 3.00 5.00 0.27 0.00 32.50 39.25 0.61 30.75 1.59 0.78 0.41 0.55 0.00 0.00 0.01 0.00 4.00 0.79 0.00 1.36 0.01 0.00 56.50 109.14 0.43 0.58 83.00 0.00 16.00 18.00 46.00 18.00 39.00 12.00 20.00 15.00 1.31 0.57/15.00

#### Sample 70:

15.63 9.38 2.88 4.75 0.63 0.00 38.38 40.00 0.57 31.25 1.68 0.78 1.70 1.13 0.13 0.21 0.02 0.00 10.27 1.71 0.00 1.64 0.04 0.00 58.25 260.21 0.43 0.66 150.00 0.28 14.00 19.00 64.00 17.00 50.00 13.00 71.00 18.00 1.04 1.15 9.00

# Sample 71:

13.50 9.00 3.00 4.75 0.41 0.00 33.50 36.50 0.57 28.88 1.51 0.79 1.43 0.57 0.00 0.21 0.04 0.00 56.57 2.29 0.00 1.27 0.04 0.00 54.88 41.27 0.41 0.61 47.00 0.00 12.00 16.00 17.00 8.00 24.00 11.00 18.00 11.00 0.86 0.92 14.00

#### Sample 72:

15.13 9.50 3.00 4.75 0.66 0.03 38.00 39.50 0.57 32.00 1.59 0.81 1.27 0.29 0.00 0.21 0.02 0.00 22.00 2.00 0.00 0.57 0.01 0.00 63.00 161.71 0.39 0.60 228.00 0.55 15.00 23.00 96.00 16.00 186.00 19.00 184.00 22.00 0.62 1.18 4.00

# Sample 73:

14.38 8.75 3.00 4.75 0.59 0.05 36.25 38.75 0.57 31.38 1.66 0.81 0.27 0.79 0.00 0.21 0.04 0.02 15.07 10.21 0.00 5.13 0.03 0.00 60.25 111.93 0.38 0.60 115.00 0.29 11.00 18.00 50.00 12.00 57.00 12.00 64.00 16.00 0.88 1.12 6.00

# Sample 74:

15.00 8.63 3.00 4.38 0.67 0.05 40.13 40.63 0.59 32.38 1.74 0.80 0.57 0.27 0.00 0.55 0.04 0.02 6.98 0.55 0.00 0.55 0.01 0.00 65.38 42.84 0.36 0.61 112.00 0.41 16.00 20.00 60.00 16.00 101.00 17.00 113.00 18.00 0.66 1.24 4.00

# Sample 75:

15.25 9.75 3.00 4.13 0.55 0.00 33.63 40.50 0.59 31.00 1.58 0.77 1.64 1.07 0.00 0.13 0.03 0.00 10.55 2.29 0.00 0.57 0.06 0.00 57.13 7.84 0.44 0.59 51.00 0.24 16.00 17.00 32.00 12.00 33.00 12.00 16.00 14.00 1.19 0.59 20.00

### Sample 76:

15.50 10.38 2.75 4.00 0.56 0.03 39.88 41.00 0.54 33.00 1.50 0.81 2.00 0.55 0.21 0.57 0.02 0.00 7.55 2.57 0.00 1.14 0.02 0.00 55.13 79.55 0.47 0.72 55.00 0.00 16.00 18.00 14.00 11.00 31.00 16.00 63.00 15.00 0.39 1.75 5.00

# Sample 77:

14.00 9.63 2.88 4.50 0.51 0.00 37.25 40.00 0.57 32.13 1.91 0.80 0.86 0.27 0.13 0.29 0.01 0.00 16.50 0.86 0.00 0.70 0.04 0.00 54.00 49.14 0.52 0.69 42.00 0.00 14.00 16.00 29.00 13.00 16.00 15.00 24.00 16.00 1.26 1.04 14.00

#### Sample 78:

16.38 9.50 3.00 5.00 0.57 0.03 35.88 39.88 0.56 32.13 1.73 0.81 2.55 0.57 0.29 0.00 0.03 0.00 11.84 0.98 0.00 0.41 0.03 0.00 55.88 126.13 0.46 0.64 89.00 0.18 9.00 18.00 35.00 13.00 41.00 13.00 38.00 14.00 0.92 1.00 11.00

#### Sample 79:

14.63 8.00 2.88 5.00 0.47 0.08 39.25 38.38 0.58 30.38 1.86 0.79 1.13 1.43 0.13 0.00 0.02 0.01 8.79 1.41 0.00 0.55 0.07 0.00 47.25 313.36 0.48 0.83 49.00 0.67 17.00 20.00 39.00 16.00 39.00 14.00 43.00 15.00 0.97 1.07 11.00

### Sample 80:

15.88 8.88 3.00 4.25 0.56 0.06 37.75 39.88 0.58 32.50 1.80 0.82 1.55 0.41 0.00 0.21 0.03 0.01 8.21 0.98 0.00 0.29 0.04 0.00 68.50 119.71 0.36 0.55 71.00 0.18 19.00 20.00 43.00 16.00 35.00 15.00 26.00 15.00 1.24 0.75 7.00

### Sample 81:

15.25 8.88 3.00 4.63 0.60 0.11 39.38 38.88 0.59 32.13 1.73 0.83 0.21 0.41 0.00 0.27 0.03 0.01 31.41 1.55 0.00 0.41 0.02 0.00 57.13 267.84 0.42 0.69 162.00 0.63 14.00 16.00 38.00 13.00 79.00 16.00 87.00 15.00 0.56 1.28 9.00

#### Sample 82:

14.38 9.88 3.13 4.63 0.54 0.06 42.38 38.75 0.58 31.88 1.47 0.82 0.84 0.98 0.13 0.27 0.01 0.01 32.84 1.64 0.00 0.98 0.04 0.00 63.38 179.13 0.38 0.68 171.00 0.68 16.00 18.00 49.00 15.00 109.00 18.00 46.00 15.00 0.72 0.68 6.00

# Sample 83:

13.63 8.50 3.00 4.50 0.48 0.05 43.50 37.63 0.58 31.38 1.63 0.83 0.84 0.86 0.00 0.57 0.01 0.02 9.14 1.13 0.00 0.55 0.06 0.00 61.88 188.98 0.36 0.70 149.00 0.44 12.00 18.00 59.00 14.00 75.00 15.00 77.00 15.00 0.84 1.09 6.00

#### Sample 84:

14.75 8.75 2.50 4.63 0.76 0.10 39.75 39.00 0.60 31.00 1.69 0.80 0.79 0.21 0.29 0.27 0.06 0.08 42.21 4.00 0.00 1.71 0.01 0.00 63.38 67.70 0.37 0.63 217.00 0.34 12.00 18.00 77.00 14.00 102.00 17.00 137.00 14.00 0.73 1.30 3.00

#### Sample 85:

14.88 8.63 2.75 4.75 0.62 0.00 37.13 38.88 0.58 30.75 1.74 0.79 1.84 1.13 0.21 0.21 0.05 0.00 36.98 4.13 0.00 3.36 0.05 0.00 53.50 72.86 0.44 0.69 241.00 0.43 12.00 17.00 68.00 15.00 114.00 14.00 158.00 14.00 0.60 1.39 3.00

# Sample 86:

15.88 9.00 3.00 4.25 0.53 0.05 38.25 38.75 0.57 32.13 1.78 0.83 1.84 0.86 0.00 0.50 0.02 0.01 17.36 1.36 0.00 1.55 0.05 0.00 66.63 211.41 0.37 0.57 187.00 0.58 12.00 19.00 88.00 14.00 72.00 13.00 130.00 15.00 0.91 1.34 8.00

# Sample 87:

15.13 9.88 2.75 3.88 0.53 0.03 42.13 39.88 0.57 32.50 1.56 0.82 0.41 1.55 0.21 0.41 0.01 0.01 28.70 0.70 0.00 0.29 0.05 0.00 63.00 136.00 0.40 0.67 143.00 0.32 13.00 18.00 65.00 15.00 82.00 15.00 72.00 15.00 0.89 0.99 6.00

# Sample 88:

15.00 9.63 3.00 4.38 0.47 0.00 41.25 39.13 0.57 32.00 1.57 0.82 1.71 0.84 0.00 0.55 0.02 0.00 15.93 0.98 0.00 0.86 0.05 0.00 68.00 183.43 0.36 0.61 187.00 0.49 11.00 16.00 143.00 13.00 191.00 19.00 296.00 17.00 0.68 1.41 3.00

#### Sample 89:

12.38 9.63 2.75 4.13 0.75 0.00 37.63 35.50 0.56 28.13 1.29 0.79 1.70 0.84 0.21 0.41 0.05 0.00 12.84 2.00 0.00 0.98 0.02 0.00 59.63 97.41 0.37 0.63 57.00 0.19 12.00 16.00 33.00 13.00 32.00 14.00 37.00 13.00 0.97 1.09 6.00

# Sample 90:

15.25 9.75 3.00 5.00 0.45 0.08 43.38 38.75 0.58 29.88 1.57 0.77 2.21 0.79 0.00 0.00 0.02 0.04 11.98 3.36 0.00 0.70 0.04 0.00 54.38 145.98 0.46 0.80 108.00 0.00 14.00 17.00 32.00 12.00 44.00 13.00 45.00 12.00 0.79 1.12 13.00

### Sample 91:

13.75 7.75 3.25 4.13 0.97 0.00 35.25 36.25 0.55 30.88 1.79 0.85 1.07 0.50 0.50 0.70 0.01 0.00 17.07 1.36 0.00 1.55 0.08 0.00 42.75 143.64 0.50 0.82 45.00 0.56 13.00 13.00 24.00 9.00 35.00 9.00 83.00 10.00 0.51 1.75 9.00

#### Sample 92:

12.75 7.88 3.25 4.88 0.84 0.00 33.88 35.63 0.55 30.38 1.66 0.85 1.64 0.98 0.21 0.13 0.03 0.00 14.13 1.13 0.00 1.13 0.16 0.00 44.63 51.13 0.46 0.76 122.00 0.70 11.00 14.00 42.00 13.00 51.00 12.00 52.00 10.00 0.87 1.08 4.00

#### Sample 93:

12.75 8.13 2.88 4.38 0.89 0.00 31.13 36.00 0.57 30.38 1.59 0.84 1.64 0.70 0.13 0.27 0.01 0.00 16.13 0.57 0.00 0.27 0.08 0.00 52.00 61.14 0.40 0.60 65.00 0.17 13.00 14.00 23.00 10.00 22.00 10.00 29.00 12.00 0.93 1.18 20.00

# Sample 94:

14.63 8.00 3.00 4.38 0.54 0.09 31.75 39.13 0.58 30.75 1.84 0.79 0.27 0.29 0.00 0.27 0.01 0.03 11.36 0.70 0.00 0.79 0.03 0.00 56.25 79.07 0.40 0.56 126.00 0.36 12.00 17.00 24.00 13.00 28.00 13.00 62.00 14.00 0.63 1.63 6.00

#### Sample 95:

13.88 8.13 3.13 4.75 0.42 0.00 30.50 38.13 0.57 30.63 1.72 0.80 1.27 0.41 0.13 0.21 0.02 0.00 37.71 1.55 0.00 1.98 0.07 0.00 55.25 17.07 0.40 0.55 98.00 0.21 12.00 17.00 30.00 11.00 39.00 12.00 45.00 12.00 0.79 1.18 9.00

# Sample 96:

15.00 8.50 3.13 4.75 0.53 0.03 34.00 38.88 0.56 31.25 1.78 0.80 1.43 0.86 0.13 0.50 0.02 0.00 6.86 0.70 0.00 0.50 0.06 0.00 56.25 67.36 0.42 0.60 100.00 0.32 12.00 16.00 37.00 12.00 38.00 13.00 55.00 14.00 0.85 1.27 8.00

# Sample 97:

14.63 10.00 2.88 4.13 0.71 0.09 35.50 40.63 0.58 32.00 1.39 0.79 2.27 0.57 0.13 0.41 0.05 0.01 7.43 0.27 0.00 0.29 0.06 0.00 53.00 55.43 0.48 0.67 147.00 0.24 10.00 16.00 24.00 12.00 54.00 12.00 53.00 13.00 0.55 1.21 6.00

# Sample 98:

15.63 9.75 3.13 4.50 0.45 0.36 39.38 39.88 0.58 31.38 1.61 0.79 0.84 0.50 0.13 0.29 0.00 0.07 29.70 1.55 0.00 1.13 0.03 0.00 54.25 103.64 0.47 0.73 122.00 0.37 15.00 18.00 54.00 14.00 81.00 14.00 115.00 17.00 0.65 1.38 5.00

### Sample 99:

13.13 8.00 3.00 4.75 0.53 0.06 31.25 35.38 0.56 30.25 1.68 0.86 2.13 1.14 0.00 0.21 0.04 0.01 14.21 1.70 0.00 1.07 0.15 0.00 47.25 82.79 0.45 0.66 126.00 0.46 10.00 13.00 44.00 12.00 109.00 12.00 355.00 15.00 0.26 2.10 5.00

#### Sample 100:

13.88 8.13 2.88 4.50 0.80 0.05 34.38 36.50 0.58 30.25 1.72 0.83 0.98 0.41 0.13 0.57 0.05 0.02 13.41 1.43 0.00 0.79 0.03 0.00 45.00 128.00 0.49 0.76 135.00 0.64 11.00 16.0 90.00 16.00 129.00 14.00 319.00 16.00 0.50 1.78 6.00

# Sample 101:

14.88 8.00 2.75 4.13 0.81 0.11 34.88 37.63 0.59 31.63 1.87 0.84 0.70 0.57 0.21 0.41 0.02 0.01 4.98 0.84 0.00 0.84 0.03 0.00 45.25 184.50 0.51 0.77 103.00 0.24 13.00 16.00 41.00 14.00 45.00 15.00 17.00 0.94 1.03 6.00

### Sample 102:

13.75 9.13 2.75 4.38 0.44 0.05 31.50 36.25 0.61 29.50 1.52 0.82 2.79 0.70 0.21 0.27 0.04 0.01 8.86 15.64 0.00 1.71 0.06 0.01 50.75 101.07 0.45 0.62 148.00 0.40 14.00 17.00 62.00 14.00 122.00 14.00 128.00 15.00 0.60 1.23 6.00

# Sample 103:

13.50 9.00 2.88 4.38 0.40 0.03 31.25 37.13 0.58 30.25 1.55 0.82 0.86 2.00 0.13 0.27 0.01 0.00 5.93 1.84 0.00 0.79 0.14 0.00 57.13 110.41 0.39 0.55 74.00 0.50 14.00 15.00 42.00 13.00 46.00 13.00 43.00 13.00 0.96 0.98 21.00

# Sample 104:

13.75 9.00 3.00 4.75 0.29 0.03 30.00 38.00 0.57 31.00 1.54 0.82 1.07 0.57 0.00 0.21 0.03 0.00 20.86 1.14 0.00 1.71 0.04 0.00 51.88 127.27 0.44 0.58 124.00 0.39 12.00 17.00 20.00 16.00 68.00 12.00 72.00 13.00 0.38 1.35 17.00

#### Sample 105:

12.75 8.63 2.88 4.50 0.14 0.00 25.63 35.00 0.60 30.25 1.49 0.86 0.21 0.84 0.13 0.29 0.01 0.00 5.70 0.57 0.00 0.21 0.03 0.00 43.50 24.57 0.49 0.59 31.00 0.42 12.00 13.00 22.00 7.00 23.00 8.00 31.00 5.00 0.87 1.22 11.00

#### Sample 106:

13.75 9.13 3.00 4.38 0.22 0.00 29.88 36.38 0.56 30.63 1.42 0.84 3.07 0.41 0.00 0.55 0.03 0.00 22.70 1.13 0.00 1.13 0.04 0.00 49.38 27.41 0.50 0.61 115.00 0.72 11.00 13.00 45.00 11.00 55.00 12.00 23.00 10.00 1.10 0.56 9.00

#### Sample 107:

11.75 8.38 3.00 4.63 0.23 0.00 26.88 35.88 0.58 30.00 1.43 0.84 1.07 1.13 0.00 0.27 0.04 0.00 4.41 0.41 0.00 0.57 0.05 0.00 48.50 37.43 0.41 0.55 56.00 0.00 14.00 15.00 28.00 10.00 25.00 10.00 33.00 11.00 0.98 1.15 11.00

#### Sample 108:

13.38 8.75 3.00 4.13 0.27 0.00 25.13 37.63 0.53 30.63 1.54 0.81 0.84 0.50 0.00 0.41 0.02 0.00 25.84 1.13 0.00 0.84 0.03 0.00 47.00 59.43 0.47 0.53 72.00 0.00 12.00 12.00 27.00 11.00 37.00 10.00 37.00 9.00 0.80 1.10 13.00

#### Sample 109:

13.38 8.25 2.75 3.75 0.44 0.00 29.63 36.38 0.57 30.63 1.64 0.84 0.84 0.79 0.21 0.50 0.04 0.00 12.27 0.84 0.00 0.55 0.06 0.00 47.25 62.79 0.46 0.63 70.00 0.50 13.00 14.00 32.00 8.00 36.00 8.00 45.00 11.00 0.85 1.19 14.00

## Sample 110+

13.00 9.63 2.88 438 0.55 0.00 32.75 36.88 0.56 30.00 1.36 0.81 0.57 0.27 0.13 0.55 0.02 0.00 6.50 0.70 0.00 0.57 0.02 0.00 43.75 117.93 0.52 0.75 106.00 0.42 9.00 16.00 31.00 10.00 58.00 12.00 69.00 12.00 0.59 1.31 10.00

# Sample 111:

14.25 8.38 4.38 0.26 0.00 28.13 36.50 0.57 2.88 0.50 0.55 0.13 0.27 0.04 0.00 56.63 65.13 0.40 0.50 17.84 0.86 1.71 0.81 0.02 0.00 91.00 0.55 0.00 0.57 53.00 7.00 44.00 5.00 0.97 0.92 11.00 46.00 8.00 8.00 9.00

#### Sample 112:

3.75 1.07 27.88 37.50 0.53 13.25 8.25 2.63 0.23 0.00 0.27 0.50 0.01 0.00 16.41 0.86 1.63 0.83 0.50 56.25 63.36= 0.38 0.50 80.00 0.06 0.41 0.06 0.00 35.00 8.00 45.00 9.00 0.98 1.13 15.00 39.00 8.00 7.00 10.00

### Sample 113:

13.00 8.13 3.00 3.88 0.31 0.00 25.75 37.50 0.60 28.38 1.63 0.76 0.57 0.98 0.00 0.41 0.06 0.00 13.36 0.57 0.00 12.27 0.07 0.01 57.13 30.41 0.37 0.45 75.00 0.51 9.00 12.00 24.00 6.00 33.00 7.00 38.00 5.00 0.76 1.20 13.00

#### Sample 114:

15.13 8.75 3.00 4.13 0.09 0.00 27.38 38.13 0.57 31.88 1.77 0.84 0.70 1.36 0.00 0.13 0.01 0.00 13.70 0.70 0.00 0.41 0.11 0.00 47.25 101.07 0.51 0.58 104.00 0.72 9.00 14.00 46.00 7.00 60.00 10.00 65.00 11.00 0.81 1.14 13.00

#### Sample 115:

13.25 8.63 3.00 4.50 0.40 0.00 31.00 36.25 0.58 30.75 1.56 0.85 1.36 0.84 0.00 0.29 0.04 0.00 12.29 0.50 0.00 1.93 0.07 0.00 43.25 62.50 0.51 0.72 47.00 0.47 11.00 12.00 34.00 10.00 37.00 6.00 42.00 9.00 0.90 1.12 11.00

#### Sample 116:

14.38 7.63 3.00 3.63 0.43 0.00 28.88 36.88 0.62 30.00 1.90 0.81 0.55 0.55 0.00 0.27 0.03 0.00 4.13 2.70 0.00 2.00 0.04 0.00 46.50 159.14 0.47 0.62 29.00 0.00 11.00 11.00 25.00 7.00 33.00 9.00 24.00 9.00 0.91 0.88 16.00

#### Sample 117:

13.50 9.50 3.00 4.50 0.39 0.00 31.88 39.75 0.58 33.38 1.44 0.84 0.57 0.86 0.00 0.29 0.04 0.00 15.27 3.36 0.00 0.84 0.04 0.00 52.75 109.64 0.44 0.60 160.00 0.53 8.00 14.00 65.00 11.00 50.00 11.00 52.00 9.00 1.17 0.93 5.00

# Sample 118:

13.25 9.00 2.75 4.13 0.51 0.00 33.75 39.13 0.59 33.50 1.48 0.86 0.50 0.29 0.21 0.41 0.03 0.00 10.50 0.98 0.00 0.57 0.02 0.00 48.50 82.00 0.46 0.70 102.00 0.61 10.00 13.00 22.00 7.00 41.00 11.00 52.00 8.00 0.57 1.36 13.00

# Sample 119:

11.88 8.88 3.00 4.63 0.51 0.03 29.88 37.50 0.58 31.50 1.34 0.84 0.41 0.41 0.00 0.27 0.02 0.00 13.84 0.57 0.00 1.71 0.01 0.00 45.50 99.71 0.46 0.66 136.00 0.46 8.00 14.00 40.00 9.00 43.00 10.00 54.00 10.00 0.88 1.18 12.00

# Sample 120:

14.38 9.63 2.88 4.25 0.52 0.00 32.50 38.00 0.57 31.25 1.50 0.82 0.55 0.84 0.13 0.50 0.03 0.00 10.57 0.86 0.00 0.79 0.01 0.00 57.88 138.13 0.41 0.56 145.00 0.79 8.00 17.00 50.00 11.00 60.00 10.00 70.00 10.00 0.83 1.17 11.00

#### Sample 121:

13.75 9.00 3.00 4.38 0.46 0.00 31.75 37.88 0.58 30.50 1.54 0.81 1.07 0.57 0.00 0.27 0.03 0.00 31.36 3.55 0.00 0.86 0.04 0.00 55.25 119.93 0.41 0.57 37.00 0.54 10.00 11.00 28.00 8.00 44.00 7.00 37.00 6.00 0.77 1.02 11.00

# Sample 122:

15.38 9.50 3.00 4.88 0.69 0.00 33.00 38.50 0.59 30.63 1.62 0.80 0.55 0.29 0.00 0.13 0.03 0.00 10.57 0.86 0.00 0.55 0.02 0.00 61.00 238.57 0.41 0.54 46.00 0.54 16.00 18.00 54.00 13.00 55.00 15.00 51.00 12.00 1.01 0.96 12.00

# Sample 123:

14.25 8.88 3.00 4.00 0.62 0.00 37.25 38.75 0.59 31.88 1.61 0.82 0.50 0.41 0.00 0.57 0.02 0.00 19.93 1.64 0.00 0.13 0.02 0.00 47.88 132.41 0.48 0.78 72.00 0.00 15.00 14.00 30.00 10.00 30.00 10.00 37.00 12.00 0.93 1.14 19.00

# Sample 124:

14.50 8.75 3.13 4.25 0.42 0.00 32.75 39.25 0.62 31.63 1.67 0.81 0.86 1.07 0.13 0.50 0.08 0.00 52.79 1.64 0.00 1.70 0.03 0.00 57.38 95.70 0.41 0.57 78.00 0.00 14.00 17.00 23.00 11.00 28.00 11.00 38.00 12.00 0.78 1.28 11.00

# Sample 125:

0.28 0.00 31.75 39.88 0.60 4.63 15.00 8.25 3.00 0.55 0.03 0.00 38.79 0.98 1.86 0.80 1.36 0.00 1.43 92.00 0.05 0.00 0.79 0.14 0.00 56.75 2**49.**64 0.41 0.56 15.00 30.00 8.00 39.00 11.00 36.00 9.00 0.86 1.03 9.00 14.00

# Sample 126:

13.00 8.50 2.75 4.75 0.36 0.00 26.13 36.75 0.55 31.13 1.54 0.85 0.86 0.57 0.21 0.21 0.05 0.00 5.84 0.79 0.00 0.70 0.03 0.00 52.25 98.07 0.41 0.50 11.00 0.45 12.00 12.00 30.00 9.00 35.00 11.00 40.00 10.00 0.86 1.14 8.00

#### Sample 127:

12.88 7.63 2.88 4.13 0.40 0.00 28.13 37.25 0.57 31.38 1.73 0.84 0.98 1.13 0.13 0.41 0.05 0.00 12.41 0.21 0.00 0.27 0.12 0.00 48.00 90.00 0.43 0.59 87.00 0.29 13.00 15.00 34.00 10.00 40.00 11.00 16.00 13.00 1.13 0.53 9.00

# Sample 128:

13.13 8.38 2.88 4.13 0.54 0.00 31.50 37.13 0.57 31.75 1.58 0.86 0.41 0.84 0.13 0.70 0.02 0.00 8.29 0.13 0.00 0.50 0.02 0.00 42.38 127.70 0.51 0.74 156.00 0.38 11.00 14.00 58.00 12.00 82.00 12.00 105.00 13.00 0.71 1.29 6.00

# Sample 129:

13.25 8.63 3.00 4.38 0.51 0.00 31.00 37.38 0.61 29.63 1.55 0.79 1.36 0.84 0.00 0.27 0.05 0.00 24.00 0.84 0.00 0.55 0.04 0.00 47.75 31.93 0.46 0.65 52.00 0.56 10.00 11.00 18.00 7.00 22.00 8.00 22.00 7.00 0.87 1.06 11.00

#### Sample 130:

12.50 7.50 2.38 4.13 0.41 0.00 24.25 34.50 0.60 27.63 1.70 0.80 0.86 1.14 0.27 0.98 0.09 0.00 24.50 0.86 0.00 1.13 0.08 0.00 44.00 245.14 0.45 0.55 110.00 0.46 9.00 14.00 67.00 9.00 60.00 10.00 92.00 11.00 0.92 1.26 5.00

#### Sample 131:

4.50 0.39 0.00 28.13 35.88 0.56 12.63 8.00 3.00 0.00 0.29 0.02 0.00 4.41 1.27 1.59 0.87 0.29 0.55 34.50 11.71 0.60 0.82 14.00 0.00 0.00 0.79 0.03 0.00 28.00 7.00 49.00 10.00 49.00 8.00 0.67 8.00 11.00 8.00

# Sample 132:

13.50 8.13 2.75 4.00 0.42 0.00 30.00 38.25 0.59 30.75 1.69 0.80 0.86 1.27 0.21 0.57 0.04 0.00 25.71 0.79 0.00 0.21 0.07 0.00 48.88 127.27 0.44 0.61 96.00 0.65 12.00 14.00 25.00 10.00 25.00 9.00 29.00 9.00 0.95 1.10 14.00

#### Sample 133:

11.63 8.38 2.38 0.24 0.00 3.63 24.38 34.50 0.54 30.25 1.98 1.42 0.88 1.41 0.27 0.27 0.05 0.00 13.98 0.57 0.00 43.88 9.27 0.21 0.10 0.00 0.46 0.56 25.00 0.00 10.00 17.00 7.00 28.00 5.00 9.00 39.00 8.00 0.61 1.39 11.00

#### Sample 134:

12.63 8.38 0.39 0.00 24.88 36.00 0.58 3.00 3.63 0.57 1.52 0.82 0.84 0.55 0.00 0.27 0.02 0.00 5.27 50.63 72.84 0.41 0.49 0.00 0.57 0.03 0.00 76.00 0.46 8.00 10.00 30.00 6.00 30.00 6.00 34.00 7.00 0.96 1.09 11.00

#### Sample 135:

13.13 7.50 3.00 4.50 0.39 0.03 25.88 35.38 0.55 29.38 1.78 0.83 0.41 0.86 0.00 0.29 0.02 0.01 2.13 2.84 0.00 1.70 0.07 0.00 50.50 35.14 0.41 0.51 22.00 0.00 12.00 12.00 20.00 10.00 30.00 9.00 29.00 9.00 0.76 1.10 16.00

# Sample 136:

30.63 39.13 0.61 30.75 14.50 8.25 4.25 0.56 0.00 2.88 7.98 0.98 1.78 0.79 0.03 0.00 1.07 0.13 0.21 0.57 0.79 0.06 0.00 57.25 243.64 0.40 0.53 115<sub>0</sub>00 0.55 0.00 1.22 0.59 13.00 16.00 64.00 11.00 62.00 10.00 31.00 8.00 9.00

# Sample 137:

13.00 7.63 2.88 4.00 0.53 0.00 31.50 37.50 0.58 30.25 1.72 0.81 3.14 0.27 0.13 0.29 0.06 0.00 48.00 0.86 0.00 0.50 0.09 0.00 49.63 91.98 0.42 0.63 92.00 0.60 10.00 13.00 27.00 8.00 42.00 8.00 41.00 10.00 0.74 1.12 7.00

# Sample 138:

13.50 8.88 0.62 0.00 34.88 **35.**50 0.58 29.25 3.00 4.13 1.53 0.83 0.41 0.06 0.00 76.13 3.43 0.41 1.14 0.00 0.00 0.79 0.02 0.00 56.75 183.36 0.39 0.61 283.00 0.74 8.00 17.00 129.00 14.00 176.00 15.00 155.00 16.00 0.84 3.00

# Sample 139:

14.00 7.88 3.00 4.63 0.66 0.00 31.38 36.00 0.57 30.13 1.78 0.84 1.43 0.41 0.00 0.27 0.03 0.00 8.27 1.71 0.00 0.98 0.02 0.00 45.13 126.70 0.48 0.70 52.00 0.44 12.00 14.00 47.00 8.00 66.00 9.00 92.00 10.00 0.69 1.35 10.00

### Sample 140:

13.75 8.00 2.75 4.25 0.81 0.00 33.25 36.75 0.58 30.38 1.74 0.83 1.36 1.14 0.21 0.50 0.03 0.00 27.64 3.93 0.00 3.41 0.05 0.00 49.25 159.36 0.44 0.68 192.00 0.83 8.00 15.00 70.00 14.00 70.00 11.00 86.00 11.00 0.93 1.14 6.00

# Sample 141:

13.38 6.88 2.75 5.00 0.72 0.00 32.13 34.25 0.56 29.25 1.96 0.85 2.27 0.41 0.21 0.00 0.03 0.00 23.84 0.79 0.00 2.21 0.08 0.00 55.63 90.55 0.36 0.58 112.00 0.62 8.00 12.00 29.00 7.00 48.00 7.00 47.00 9.00 0.70 1.14 14.00

#### Sample 142:

13.38 6.88 2.75 3.50 0.50 0.00 27.50 35.38 0.63 29.50 1.96 0.83 1.98 0.41 0.21 0.29 0.02 0.00 15.14 0.84 0.00 0.57 0.08 0.00 49.00 20.29 0.41 0.56 23.00 0.00 8.00 9.00 18.00 6.00 23.00 6.00 31.00 5.00 0.75 1.29 17.00

#### Sample 143:

14.13 7.00 3.00 3.75 0.37 0.00 23.25 37.38 0.62 29.75 2.04 0.80 0.41 0.57 0.00 0.50 0.07 0.00 5.07 0.27 0.00 1.07 0.07 0.00 50.63 55.13 0.42 0.46 29.00 0.52 12.00 14.00 29.00 8.00 41.00 10.00 35.00 9.00 0.83 1.00 14.00

#### Sample 144:

15.38 7.50 3.00 3.75 0.40 0.00 27.75 36.63 0.57 30.38 2.07 0.83 0.27 0.57 0.00 0.50 0.07 0.00 15.93 0.55 0.00 1.13 0.06 0.00 47.88 145.27 0.48 0.58 89.00 0.71 7.00 12.00 45.00 11.00 52.00 11.00 52.00 10.00 0.91 1.05 9.00

### Sample 145:

14.25 8.75 3.00 4.38 0.35 0.00 30.38 38.00 0.57 30.38 1.64 0.80 1.07 0.21 0.00 0.55 0.02 0.00 18.27 0.86 0.00 15.13 0.03 0.01 48.63 142.27 0.47 0.62 117.00 0.40 9.00 16.00 31.00 11.00 37.00 9.00 36.00 9.00 0.89 1.04 11.00

# Sample 146;

12.75 8.63 3.00 4.50 0.37 0.00 29.75 36.00 0.58 0.00 0.57 0.27 1.48 0.84 0.21 0.03 0.00 19.36 1.14 0.00 0.50 0.01 0.00 43.75 29.64 0.49 0.68 74.00 0.45 12.00 13.00 26.00 8.00 40.00 8.00 45.00 8.00 0.70 1.22 11.00

#### Sample 147:

0.31 12.38 8.13 3.00 4.13 0.00 30.13 36.00 0.57 1.53 0.86 0.41 0.00 0.70 1.13 0.01 0.00 14.70 0.57 0.00 0.13 0.03 0.00 42.88 8.41 0.48 0.70 67.00 0.51 12.00 15.00 12.00 9.00 19.00 9.00 31.00 11.00 0.58 1.50 10.00

## Sample 148:

12.75 8.25 3.00 4.25 0.41 0.00 28.63 38.25 0.56 32.38 1.56 0.85 0.50 0.79 0.00 0.21 0.02 0.00 10.84 0.79 0.00 0.27 0.04 0.00 42.50 62.29 0.49 0.67 67.00 0.72 11.00 13.00 30.00 7.00 31.00 10.00 32.00 10.00 0.97 1.03 10.00

#### Sample 149:

13.00 9.25 2.88 4.13 0.46 0.00 30.25 37.13 0.60 31.50 1.41 0.85 0.86 0.50 0.13 0.41 0.03 0.00 11.07 0.98 0.00 0.29 0.03 0.00 49.25 78.21 0.45 0.61 86.00 0.33 14.00 15.00 74.00 15.00 90.00 14.00 94.00 16.00 0.86 1.09 5.00

#### Sample 150:

11.88 8.50 4.13 0.46 0.00 25.25 36.50 0.58 2.75 1.43 0.85 0.21 0.41 2.29 0.03 0.00 6.21 0.13 1.43 58.00 0.40 0.00 1.27 0.07 0.00 38.88 61.55 0.52 0.65 10.00 15.00 18.00 6.00 23.00 8.00 22.00 6.00 0.86 9.00

# Sample 151:

30.75 36.75 0.57 13.38 8.63 30.75 3.00 4.25 0.41 0.00 1.58 0.84 0.84 0.03 0.00 13.07 1.07 1.13 0.00 0.50 44.13 74.70 0.50 0.00 0.50 0.10 0.70 59.00 0.42 0.00 8.00 12.00 18.00 5.00 33.00 8.00 25.00 6.00 0.71 0.99 8.00

# Sample 152:

13.13 9.38 2.88 4.63 0.43 0.00 31.38 36.25 0.58 30.63 1.41 0.85 0.41 1.13 0.13 0.27 0.01 0.00 14.55 0.50 0.00 0.55 0.03 0.00 44.38 77.13 0.51 0.71 61.00 0.31 10.00 11.00 14.00 8.00 15.00 10.00 17.00 10.00 0.91 1.11 10.00

# Sample 153:

32.38 11.13 13.00 3.00 4.50 1.00 0.00 38.38 37.88 0.56 0.00 0.29 0.57 0.86 0.86 0.41 0.00 0.00 0.98 9.41 0.00 40.88 121.27 0.59 0.94 0.84 0.01 88.00 0.48 0.00 11.00 26.00 10.00 36.00 9.00 59.00 9.00 8.00 0.64 6.00

#### Sample 154:

12.75 8.25 3.00 4.25 0.27 0.00 29.25 35.13 0.53 0.50 0.50 1.56 0.88 24.50 0.70 0.00 0.21 0.01 0.00 0.86 0.03 0.00 0.00 44.50 78.00 0.47 0.66 138.00 0.45 8.00 15.00 61.00 11.00 64.00 11.00 71.00 10.00 0.93 1.09 7.00

#### Sample 155:

11.38 11.75 2.75 4.25 1.00 0.00 39.00 37.38 0.57 30.88 0.50 0.21 0.00 0.00 7.14 0.27 0.97 0.83 0.55 0.21 0.00 0.98 0.01 0.00 43.88 51.55 0.53 0.89 67.00 0.31 12.00 27.00 9.00 34.00 8.00 41.00 9.00 0.79 1.21 9.00 6.00

#### Sample 156:

30.25 35.50 0.54 12.75 9.50 4.50 0.48 0.00 30.75 3.00 0.86 0.01 0.00 10.79 2.00 1.35 0.87 1.07 0.00 0.57 0.00 41.13 48.70 0.54 0.00 0.50 0.74 97.00 0.21 0.02 7.00 12.00 26.00 6.00 32.00 8.00 41.00 11.00 0.79 1.24 10.00

# Sample 157:

4.88 0.41 0.00 28.75 36.50 0.56 31.13 12.75 8.75 2.88 0.50 0.13 0.13 0.01 0.00 18.79 1.14 1.47 0.85 0.50 0.00 93.00 0.53 0.00 42.00 28.00 0.51 0.68 0.41 0.02 29.00 9.00 32.00 7.00 0.97 1.07 8.00 12.00 29.00 8.00 9.00

#### Sample 158:

12.00 9.13 3.00 4.50 0.62 0.00 33.00 34.38 0.56 30.00 1.32 0.87 1.14 0.41 0.00 0.29 0.03 0.00 18.00 1.41 0.00 0.29 0.02 0.00 44.88 46.13 0.47 0.74 70.00 0.41 11.00 14.00 18.00 10.00 25.00 7.00 25.00 10.00 0.79 1.10 13.00

#### Sample 159:

13.75 8.38 3.00 4.25 0.49 0.00 31.00 36.75 0.57 30.88 1.65 0.84 1.07 0.27 0.00 0.21 0.03 0.00 8.29 2.79 0.00 1.27 0.02 0.00 56.50 95.14 0.39 0.55 164.00 0.57 10.00 19.00 71.00 13.00 133.00 16.00 141.00 14.00 0.62 1.23 3.00

### Sample 160:

12.38 8.25 3.13 4.50 0.34 0.05 30.63 35.00 0.55 1.52 0.85 0.50 0.29 0.02 0.02 6.27 0.55 0.13 2.86 0.04 0.00 41.25 47.36=0.50 0.74 0.00 1.07 181.00 0.81 15.00 101.00 12.00 171.00 13.00 198.00 14.00 0.64 9.00 6.00

#### Sample 161:

3.00 2.55 13.38 8.00 4.63 0.37 0.00 30.88 35.63 0.56 0.57 1.68 0.86 0.00 0.27 0.08 0.00 16.41 3.70 0.00 2.00 0.04 0.00 41.63 89.70 0.51 0.74 72.00 0.46 12.00 13.00 47.00 9.00 74.00 12.00 52.00 9.00 0.82 0.90 10.00

### Sample 162:

0.28 0.00 12.00 8.25 3.00 4.63 24.75 34.38 0.57 1.51 0.84 2.86 1.36 0.57 0.27 0.02 0.00 18.50 1.41 0.00 0.20 0.00 44.00 111.43 0.46 0.56 134.00 0.59 0.50 11.00 64.00 10.00 67.00 9.00 108.00 10.00 0.80 7.00 4.00

#### Sample 163:

11.63 6.88 23.13 33.88 0.56 2.63 4.25 0.11 0.00 1.72 0.86 1.13 0.70 1.27 2.41 0.27 0.21 0.01 0.00 0.00 0.70 62.00 0.37 0.09 0.00 34.25 32.50 0.54 0.68 16.00 7.00 25.00 8.00 8.00 9.00 20.00 6.00 0.98 1.23 12.00

#### Sample 164:

14.88 9.38 2.88 0.00 31.00 41.13 0.54 4.25 0.37 0.50 0.06 0.00 34.00 0.98 1.60 0.85 1.84 0.55 0.13 0.00 75.00 0.00 0.57 0.04 0.00 56.75 101.36 0.43 0.55 15.00 17.00 78.00 14.00 76.00 12.00 78.00 14.00 1.01 8.00

# Sample 165:

14.13 9.50 28.25 42.13 0.53 3.00 4.38 0.33 0.00 37.36 1.27 1.49 0.82 1.27 0.55 0.05 0.00 0.57 0.00 0.00 0.55 47.75 54.21 0.49 0.59 133.00 0.00 0.01 0.00 13.00 20.00 93.00 14.00 68.00 14.00 103.00 15.00 1.06 1.17 9.00

# Sample 166:

14.63 9.13 30.63 40.25 0.53 34.88 2.88 4.75 0.35 0.00 1.60 0.87 0.84 0.21 0.04 0.00 8.84 2.21 0.13 0.13 0.00 0.98 53.38 48.27 0.44 0.57 121.00 0.00 0.01 0.00 12.00 17.00 56.00 13.00 66.00 15.00 57.00 14.00 0.94 0.96 9.00

# Sample 167:

14.25 9.50 3.00 4.13 0.34 0.00 31.25 40.75 0.55 34.63 1.51 0.85 1.07 0.57 0.00 0.41 0.05 0.00 39.07 1.36 0.00 0.27 0.03 0.00 45.75 147.93 0.52 0.68 103.00 0.00 12.00 17.00 53.00 12.00 62.00 14.00 62.00 12.00 0.90 1.05 8.00

# Sample 168:

17.00 10.13 3.00 4.75 0.56 0.00 41.38 39.88 0.82 32.50 1.68 0.82 2.00 0.70 0.00 0.21 0.03 0.00 12.84 2.13 0.00 0.86 0.01 0.00 59.88 190.41 0.45 0.69 225.00 0.77 16.00 17.00 134.00 14.00 164.00 13.00 252.00 12.00 0.73 1.37 4.00

# Sample 169:

15.38 10¢00 2.88 4.75 0.47 0.00 37.25 38.88 0.59 32.13 1.54 0.83 2.27 0.57 0.13 0.21 0.01 0.00 23.36 2.13 0.00 1.27 0.01 0.00 57.75 51.64 0.44 0.65 233.00 0.69 14.00 14.00 17.00 11.00 37.00 12.00 191.00 14.00 0.21 2.34 4.00

# Sample 170:

16.50 11.13 3.00 4.13 0.53 0.00 39.50 39.00 0.57 33.00 1.49 0.85 0.29 0.41 0.00 0.41 0.01 00.00 28.00 2.29 0.00 0.29 0.01 0.00 72.13 115.55 0.38 0.55 126.00 0.40 13.00 17.00 14.00 10.00 29.00 11.00 95.00 12.00 0.30 2.07 4.00

# Sample 171:

18.25 10.50 2.75 4.63 0.62 0.00 43.63 41.00 0.62 33.25 1.75 0.81 0.79 0.86 0.21 0.27 0.08 0.00 15.41 1.43 0.00 0.50 0.03 0.00 51.63 327.13 0.56 0.85 112.00 0.63 13.00 16.00 68.00 14.00 74.00 15.00 76.00 13.00 0.94 1.05 8.00

# Sample 172:

17.38 9.88 2.63 5.00 0.87 0.00 46.13 41.50 0.61 33.63 1.76 0.81 0.27 0.41 0.27 0.00 0.05 0.00 14.98 1.14 0.00 0.84 0.01 0.00 50.13 412.98 0.54 0.92 307.00 0.83 16.00 19.00 84.00 12.00 270.00 18.00 256.00 16.00 0.41 1.26 2.00

# Sample 173:

17.50 10.38 2.88 4.50 0.53 0.00 40.75 40.63 0.62 31.75 1.69 0.78 1.14 0.27 0.13 0.29 0.01 0.00 5.36 2.27 0.00 0.50 0.03 0.00 49.63 44.27 0.56 0.82 136.00 0.71 12.00 13.00 42.00 9.00 59.00 12.00 54.00 12.00 0.81 1.05 10.00

# Sample 174:

16.25 9.88 3.00 5.00 0.30 0.00 39.13 42.75 0.58 34.63 1.65 0.81 0.79 0.13 0.00 0.00 0.02 0.00 9.84 0.50 0.00 0.55 0.02 0.00 59.13 316.13 0.44 0.66 237.00 0.50 16.00 21.00 139.00 18.00 127.00 20.00 189.00 18.00 0.92 1.25 2.00

#### Sample 175:

17.00 10.00 2.88 5.00 0.32 0.00 39.88 44.00 0.57 34.50 1.71 0.79 3.14 0.29 0.13 0.00 0.01 0.00 6.98 4.29 0.00 0.57 0.05 0.00 57.50 179.00 0.47 0.69 259.00 0.53 11.00 22.00 112.00 19.00 148.00 21.00 212.00 21.00 0.71 1.35 3.00

#### Sample 176:

15.63 9.88 3.00 4.75 0.30 0.00 37.13 41.88 0.55 34.63 1.59 0.83 0.55 0.70 0.00 0.21 0.02 0.00 15.27 1.27 0.00 0.27 0.01 0.00 58.38 299.98 0.44 0.57 182.00 0.43 12.00 20.00 83.00 17.00 130.00 17.00 235.00 17.00 0.56 1.57 2.00

#### Sample 177:

13.75 11.63 2.63 5.00 0.97 0.00 34.63 41.00 0.65 32.63 1.19 0.80 0.50 0.55 0.27 0.00 0.01 0.00 12.84 1.71 0.00 2.27 0.01 0.00 36.63 12.27 0.69 0.95 134.00 0.50 9.00 16.00 107.00 11.00 108.00 12.00 156.00 12.00 0.87 1.26 5.00

# Sample 178:

13.75 11.63 2.75 5.00 1.00 0.00 41.25 42.25 0.63 32.88 1.19 0.78 0.79 0.27 0.21 0.00 0.00 0.00 31.36 1.36 0.00 4.41 0.01 0.00 44.13 45.27 0.58 0.93 256.00 0.57 9.00 17.00 146.00 14.00 15800 13.00 216.00 17.00 0.84 1.25 5.00

# Sample 179:

12.88 12.25 2.63 5.00 1.00 0.00 39.38 41.25 0.64 32.63 1.06 0.79 0.98 1.64 0.27 0.00 0.00 0.00 21.13 4.50 0.00 1.41 0.01 0.00 44.50 32.86 0.56 0.88 241.00 0.58 13.00 18.00 220.00 15.00 218.00 16.00 256.00 15.00 0.95 1.11 4.00

#### Sample 180:

13.88 11.75 2.63 5.00 0.97 0.00 42.50 42.88 0.62 32.75 1.19 0.76 0.41 0.79 0.27 0.00 0.01 0.00 34.00 2.13 0.00 0.21 0.01 0.00 39.50 70.00 0.65 1.08 242.00 0.45 9.00 15.00 151.00 13.00 239.00 14.00 375.00 17.00 0.59 1.47 2.00

# Sample 181:

12.00 10.50 2.63 5.00 1.00 0.00 41.75 37.25 0.54 29.88 1.15 0.80 0.57 0.86 0.27 0.00 0.00 0.00 17.07 0.50 0.00 2.41 0.01 0.00 43.38 83.41 0.52 0.96 87.00 0.21 11.00 13.00 20.00 11.00 20.00 9.00 125.00 12.00 0.36 2.27 3.00

### Sample 182:

12.50 10.75 2.50 4.88 1.00 0.00 43.63 40.00 0.54 31.50 1.16 0.79 0.57 0.50 0.29 0.13 0.00 0.00 7.13 3.14 0.00 2.29 0.00 0.00 50.13 74.41 0.46 0.87 179.00 0.54 10.00 13.00 115.00 13.00 126.00 14.00 202.00 13.00 0.78 1.37 3.00

# Sample 183:

12.13 10.88 2.63 5.00 0.97 0.00 39.50 38.50 0.55 28.86 1.12 0.75 0.41 0.13 0.27 0.00 0.01 0.00 16.00 0.57 0.00 2.13 0.00 0.00 40.88 35.27 0.56 0.97 88.00 0.25 11.00 13.00 13.00 9.00 125.00 11.00 161.00 11.00 0.13 1.62 2.00

### Sample 184:

12.63 12.25 2.75 5.00 0.97 0.00 41.25 42.25 0.58 33.63 1.03 0.80 0.27 0.21 0.21 0.00 0.01 0.00 11.36 2.79 0.00 3.13 0.00 0;00 36.88 56.41 0.67 1.12 84.00 0.44 11.00 14.00 41.00 11.00 43.00 13.00 57.00 13.00 0.87 1.21 4.00

# Sample 185:

11.25 11.75 2.50 5.00 1.00 0.00 40.50 38.75 0.53 30.00 0.96 0.77 1.07 0.21 0.29 0.00 0.00 0.00 19.43 1.36 0.00 2.57 0.01 0.00 41.25 79.07 0.56 0.98 85.00 0.07 10.00 12.00 18.00 10.00 20.00 10.00 37.00 10.00 0.72 1.48 3.00

# Sample 186:

11.63 12.13 2.88 4.88 1.00 0.00 44.00 41.00 0.55 32.38 0.97 0.79 0.55 0.98 0.13 0.13 0.00 0.00 4.57 3.14 0.00 2.27 0.01 0.00 45.75 78.50 0.52 0.96 116.00 0.47 11.00 15.00 36.00 12.00 50.00 13.00 48.00 14.00 0.81 1.07 4.00

# Sample 187:

11.50 12.13 2.75 5.00 0.97 0.00 42.88 40.88 0.56 31.75 0.96 0.78 1.14 0.98 0.21 0.00 0.01 0.00 15.55 2.98 0.00 1.64 0.02 0.00 42.00 78.86 0.56 1.02 161.00 0.84 13.00 15.00 68.00 12.00 113.00 13.00 14.00 0.66 1.25 4.00

# Sample 188:

11.25 11.25 2.50 5.00 1.00 0.00 39.38 40.25 0.56 31.50 1.01 0.78 0.50 1.07 0.29 0.00 0.00 0.00 13.41 2.79 0.00 6.00 0.02 0.00 43.75 92.21 0.51 0.90 93.00 0.45 13.00 15.00 33.00 10.00 39.00 12.00 46.00 11.00 0.84 1.17 7.00

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# Sample 189:

10.50 11.63 2.75 5.00 1.00 0.00 36.63 38.25 0.56 32.38 0.92 0.85 9.29 2.27 0.21 0.00 0.00 0.00 17.70 0.50 0.00 0.27 0.02 0.00 41.00 54.29 0.54 0.89 71.00 0.18 13.00 14.00 41.00 12.00 49.00 10.00 75.00 12.00 0.75 1.36 9.00

# Sample 190:

11.25 11.38 3.00 5.00 1.00 0.00 39.88 39.63 0.54 32.63 0.99 0.82 1.07 0.27 0.00 0.00 0.00 0.00 **2.41** 1.41 0.00 0.84 0.02 0.00 44.75 41.07 0.51 0.89 83.00 0.00 11.00 14.00 35.00 12.00 40.00 10.00 42.00 13.00 0.90 1.08 8.00

#### Sample 191:

11.25 12.00 3.00 4.88 1.00 0.00 41.63 39.13 0.56 32.00 0.95 0.82 0.79 2.57 0.00 0.13 0.00 0.00 2.84 0.41 0.00 0.86 0.02 0.00 40.50 10.00 0.57 1.03 62.00 0.00 12.00 14.00 27.00 11.00 47.00 11.00 49.00 12.00 0.66 1.20 7.00

#### Sample 192:

11.88 10.75 2.63 5.00 1.00 0.00 39.13 40.00 0.58 32.00 1.12 0.80 0.41 1.07 0.27 0.00 0.00 0.00 10.98 1.43 0.00 0.00 0.03 0.00 39.13 90.70 0.58 1.00 74.00 0.00 9.00 13.00 23.00 10.00 27.00 9.00 33.00 12.00 0.83 1.19 9.00

# Sample 193:

10.13 11.13 3.00 5.00 1.00 0.00 37.75 38.00 0.55 30.06 0.91 0.81 0.70 0.98 0.00 0.00 0.00 0.00 5.07 0.57 0.00 0.98 0.01 0.00 37.63 79.70 0.56 1.00 52.00 0.00 8.00 12.00 17.00 8.00 17.00 10.00 20.00 9.00 0.94 1.11 9.00

### Sample 194:

10.88 11.13 3.00 5.00 1.00 0.00 35,88 38.50 0.54 30.63 0.98 0.80 0.41 0.41 0.00 0.00 0.00 0.00 7.84 0.29 0.00 2.84 0.01 0.00 38.50 23.71 0.57 0.93 49.00 0.20 9.00 12.00 14.00 7.00 15.00 10.00 20.00 10.00 0.86 1.22 7.00

# Sample 195:

10.63 10.50 2.88 4.88 1.00 0.00 38.00 36.88 0.55 31.25 1.01 0.85 0.27 0.29 0.13 0.13 0.00 0.00 11.43 0.41 0.00 0.21 0.00 0.00 46.75 29.64 0.45 0.81 111.00 0.26 10.00 17.00 36.00 13.00 35.00 12.00 36.00 14.00 1.01 1.01 4.00

#### Sample 196:

10.13 10.88 2.75 5.00 1.00 0.00 42.75 35.63 0.55 30.50 0.94 0.86 0.13 1.27 9.21 0.00 0.00 0.00 6.50 1.70 0.00 4.29 0.00 0.00 47.38 40.84 0.44 0.90 72.00 0.25 10.00 14.00 19.00 11.00 23.00 10.00 24.00 12.00 0.86 1.09 6.00

#### Sample 197:

10.25 10.38 2.88 5.00 0.97 0.00 36.75 37.63 0.55 31.13 0.99 0.83 0.79 0.55 0.13 0.00 0.01 0.00 32.79 2.27 0.00 3.27 0.01 0.00 46.38 83.98 0.44 0.79 122.00 0.36 11.00 15.00 48.00 11.00 51.00 11.00 62.00 12.00 0.89 1.16 6.00

#### Sample 198:

11.13 11.13 2.63 5.00 1.00 0.00 41.25 39.50 0.56 32.65 1.01 0.83 0.41 0.70 0.27 0.00 0.00 0.00 3.64 2.00 0.00 1.98 0.01 0.00 46.88 38.98 0.47 0.88 122.00 0.04 9.00 14.00 28.00 12.00 60.00 13.00 79.00 13.00 0.50 1.42 4.00

#### Sample 199:

30.63 36.25 0.57 31.38 1.00 0.00 9.75 10.00 2.75 5.00 0.00 0.00 0.00 14.27 1.93 1.14 0.21 0.98 0.87 1.36 0.02 0.00 31.50 39.14 0.63 0.97 77.00 0.62 3.41 7.00 0.01 19.00 5.00 27.00 8.00 36.00 6.00 0.70 1.32 6.00 7.00

#### Sample 200:

33.50 38.00 0.57 1.00 0.00 10.63 9.63 5.00 2.75 0.00 0.00 8.00 0.55 0.21 0.00 1.71 1.11 0.86 1.70 32.75 57.36 0.62 1.02 130.00 0.07 0.00 0.86 0.02 0.00 81.00 221.00 8.00 219.00 7.00 334.00 4.00 0.86 1.29 9.00 1.00

#### Sample 201:

1.00 0.00 37.88 36.25 0.57 12.25 11.63 2.38 4.63 1.41 0.27 0.27 0.00 0.00 6.41 2.21 1.06 0.84 1.64 49.88 40.98 0.48 0.76 98.00 0.88 0.00 0.02 0.00 1.43 73.00 6.00 84.00 6.00 109.00 8.00 0.82 7.00 4.00 8.00

# Sample 202:

10.25 11.00 3.00 5.00 1.00 0.00 35.13 36.88 0.53 32.38 0.94 0.88 0.50 1.14 0.00 0.00 0.00 0.00 11.84 0.70 0.00 0.55 0.02 0.00 44.00 40.86 0.48 0.80 70.00 0.50 7.00 9.00 25.00 6.00 37.00 7.00 50.00 6.00 0.67 1.34 7.00

# Sample 203:

11.13 10.75 2.63 4.88 1.00 0.00 36.75 35.00 0.56 30.13 1.05 0.86 0.41 1.93 0.27 0.13 0.00 0.00 9.64 1.43 0.00 1.84 0.02 0.00 48.88 108.13 0.45 0.75 110.00 0.75 6.00 9.00 101.00 9.00 147.00 9.00 236.00 8.00 0.63 1.46 3.00

### Sample 204:

35.75 39.63 0.57 34.13 0.00 0.97 10.75 11.00 2.75 5.00 10.79 1.70 0.21 0.00 0.01 0.00 0.79 0.29 0.98 0.86 104.00 0.55 40.88 1.55 0.53 0.87 0.01 0.00 0.70 0.00 111.00 6.00 0.75 1.10 116.00 8.00 76.00 6.00 6.00 8.00 3.00

## Sample 205:

11.75 10.63 2.75 4.88 0.97 0.00 39.63 40:00 0.57 34.88 1.11 0.87 0.50 0.27 0.21 0.13 0.01 0.00 5.41 0.57 0.00 0.98 0.01 0.00 42.50 68.86 0.53 0.93 125.00 0.72 6.00 10.00 123.00 8.00 183.00 8.00 254.00 7.00 0.66 1.36 3.00

# Sample 206:

11.38 10.75 2.88 4.88 0.94 0.00 36.63 40.00 0.56 34.38 1.08 0.86 2.27 1.64 0.13 0.13 0.01 0.00 23.13 0.86 0.00 1.70 0.05 0.00 42.63 42.84 0.52 0.86 62.00 0.40 6.00 7.00 40.00 6.00 78.00 8.00 91.00 7.00 0.57 1.31 4.00

# Sample 207:

5.00 0.80 0.00 29.25 37.75 0.57 10.00 10.63 2.75 0.21 0.00 0.02 0.00 1.64 1.07 0.55 0.95 0.85 0.00 40.88 118.41 0.50 0.72 68.00 0.51 0.00 0.00 0.00 0.98 37.00 4.00 49.00 6.00 0.78 1.27 5.00 30.00 5.00 7.00 7.00

# Sample 208:

5.00 0.70 0.00 33.75 36.63 0.60 10.63 9.63 2.88 0.00 0.02 0.00 14.50 2.27 0.55 0.84 0.13 1.11 0.83 30.00 4.00 32.00 5.00 38.00 5.00 0.90 1.14 5.00 7.00 0.02 0.00 36.88 10013 0.55 0.92 72.00 0.61 ್ರ೦.00 2.00 6.00

# Sample 209:

10.13 10.25 2.63 5.00 0.75 0.00 32.63 37.00 0.56 30.63 0.99 0.83 0.41 0.50 0.27 0.00 0.05 0.00 9.98 1.71 0.00 0.55 0.00 0.00 35.75 45.64 0.57 0.91 48.00 0.48 6.00 8.00 27.00 7.00 33.00 5.00 26.00 7.00 0.94 0.91 9.00

# Sample 210:

10.75 9.00 3.13 4.63 0.57 0.05 27.25 32.75 0.51 30.25 1.24 0.92 1.93 2.00 0.13 0.55 0.10 0.02 39.36 2.79 0.00 1.07 0.13 0.00 53.25 145.64 0.37 0.51 103.00 0.00 32.00 33.00 56.00 20.00 63.00 22.00 75.00 24.00 0.87 1.16 10.00

### Sample 211:

13.13 8.63 3.00 4.75 0.69 0.21 32.63 34.00 0.56 29.38 1.57 0.86 2.13 1.70 0.29 0.21 0.06 0.13 28.84 5.71 0.003.98 0.15 0.00 57.38 102.55 0.38 0.57 85.00 0.00 14.00 30.00 44.00 22.00 55.00 23.00 50.00 22.00 0.89 1.01 14.00

#### Sample 212:

10.63 9.59 3.00 4.50 0.76 0.13 28.75 32.38 0.51 30.63 1.12 0.95 0.84 0.29 0.00 0.29 0.02 0.02 26.21 5.13 0.00 3.98 0.02 0.00 33.63 10.27 0.60 0.86 41.00 0.00 12.00 16.00 30.00 19.00 35.00 18.00 62.00 16.00 0.71 1.46 8.00

# APPENDIX 2

# DATA FROM THE SURVEYS OF DISTRIBUTION

# A2.1 Data collected in the field

# TABLE A2.1

# INFORMATION RECORDED FOR EACH SITE IN SURVEY 1

Site number	Date of sample	Grid A	Grop <sup>2</sup> 1967	1966 Time of 3	ploughing' Addition of fertilizer	Addition of herbicide	Soil type4	Species present	A. powellii	A. retroflexus
2 3 4 5 6 7 8 9 1 1 1 1 1 1 1 1 2 2 2 2 2 2 2 2 2 2 2	3-67 3-67 3-67 3-67 3-67 3-67 3-67 7-7 7-7 7-7 7-8-67 7-8-67 7-8-67 7-8-67 7-8-67	572143 572143 572143 572143 572143 572143 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 572161 57		CHBWCCHW?TCCWTCBWHOBWCCCBCW	++-+++++++	++-++++++	BSBBBBBBBBBBBBBBBBBBBBBBBBBBBBBBBBBBB	+	+ + - + - + + + + + + + + + + + + + + + + + + + +	++++-++++++++++++++++

Site number Date of sample	Grid 1 reference $\frac{\text{Grop}^2}{\text{Grop}^2}$	7961	of Shing tion ilize	Addition of berbicide Boil type	Species present A. hybridus A. powellii A. retroflexus
2933333333333334444444444444455555555566666666	33449884 44728344 54844 5484 5484 5484 5486 5555 5555 565 565 565 565 565	#0#00#####00##0##0#0000000###00000##0₩0₩00#00₩0	++++++++++++++++++++++++++++++++++++++	PCGLLLOLLLACULUC CLALLELLLACULUC LACULUC LACUL	+ + - + + + + + + + + + + + + + + + + +

Site number Date of sample	Grid 1 reference Crop	f ing	fertilizer Addition of herbicide Soil type	Species present A. hybridus A. powellii A. retroflexus
74 6-9-67 75 6-9-67 76 6-9-67 77 6-9-67 78 6-9-67 79 30-8-67 80 30-8-67 81 30-8-67 82 30-8-67 83 30-8-67 84 30-8-67 85 30-8-67 86 30-8-67 87 30-8-67 88 30-8-67 88 30-8-67	415221 448249 457256 506189 485172 490175 479146 511142 493126 485115 513125	A&S + + + + + + + + + + + + + + + + + + +	+ HC + BSL - BSL + BEL + BEL + BEL + BEL + BEL + BEL + BEL + BEL + BEL	- + + + + + + + + + + + + + + + + + + +

#### 1 - Grid reference: Notes:

Reference figures give the position to the nearest 100 metres on the Universal Transverse Mercator Grid. The grid references in this table are between 200 and 050 East and between 860 and 650 North.

# 2 - Crops:

B - beans (soybeans, whitebeans, waxbeans)

C - corn

Ca - cabbage

Cu - cucumber

F - fallow O - oats

Po - potatoes

T - tobacco

To - tomatoes

W - wheat

# 3 - Time of ploughing:

A - autumn

S - spring

(continued)

#### 4 - Soil types:

A key to the abbreviations used to describe soil types is given in Table A2.3. Soil types were determined from Soil Survey Reports (Canada Department of Agriculture). Certain sites fell apparently upon the boundary of two soil zones. An interpretation of the symbols used to denote these sites is given below.

?1 - BEL and BSL?2 - BEL and GS?3 - BSL and BCL

?4 - BCL and HC

TABLE A2.2

INFORMATION RECORDED FOR EACH SITE IN SURVEY 2

Site no.	Grid <sup>2</sup>	Crop <sup>3</sup>	Species <sup>4</sup>	Site <sup>1</sup>	Grid <sup>2</sup> ref.	Crop <sup>3</sup>	Species <sup>4</sup>
13467891169a ba acbbass 333333333444480 a 16868686777788 a 22222233333333333333333333333333333	638220 645210 645210 631171 631178 579148 579148 579148 579148 579120 392028 376012 392003 357003 357003 357003 3570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 570176 5	BBBBCCBCBCBCTTBCTBFCBCBBCBBBCBBBBBBBBBB	PRRHEHPR RRR RRR RRR RRR RRR RRR RRR RRR RRR	23578911181222222233333333333444445555666666777780 235789111892222223333333333344445755666666777780	628194 628194 636183 622183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 636183 63	H B C C H B C B B B C C C B C C B B B B	PRESENTE RECEPTION OF THE PROPERTY OF THE PROP

(continued)

Site1	Grid <sup>2</sup> ref.	Crop <sup>3</sup>	Species <sup>4</sup>	Site <sup>1</sup>	Grid <sup>2</sup>	Crop <sup>3</sup>	Species <sup>4</sup>
81b 82b 84a 86a 879 96 99 99 1120b 121c 120b 121c 127a 129a 133b 135b 142a 148 166 168	44618 43819 40618 36214 33911 35113 34514 40519 36217	COBBBBBBBBCCBBBBTCBCBCCBBBBBBBBBBBBBBBB	PR R R R R R R R R R R R R R R R R R R	818 818 819 819 8110 8110 8110 8110 8110	498251 545294 571320 594326 488359 488359 478339 496337 496337 496337 496337 498279 508277 498279 508277 498279 508279 498279 508279 4982158 602308 5722308 5722308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 5732308 57323	BCCBBCCCBBBBBBBBBBBBBBBBBBBBBBBBBBBBB	PRR RPRR RPRPRRRRRRRRRRRRRRRRRRRRRRRRR

(continued)

Site <sup>1</sup>	Grid <sup>2</sup> ref.	Crop.3	Species <sup>4</sup>	Site no.	Grid <sup>2</sup> ref.	Crop <sup>3</sup>	Species <sup>4</sup>
201	398211	В	P	202	409197	В	P
203a	418187	C	PR	203b	422182	F	PR
204	419176	B	R		390148	C	PR
205b	405163	В	R	207	362121	C	PR
208a	379120	C	PR	208b	380121	В	HPR
209	400117	C	PR		392105	C	HPR
214b	394107	В	PR	210	338045	C	HPR
211a	344028	В	HPR	2116	347033	C F C	HPR
212	350034	В	HPR	213a	416058	C	HPR
213b	402062	В	HPR	215	373108	В	HPR
216 <b>a</b>	423062	C	PR	216b	432068	В	HPR
217	350089	C	PR	218	415102	В	R
219	390129	В	PR		436173	Ö	HPR
221b	443180	T	HR	222	470195	B	PR
223a	480204	C	P	223b	496218	B B B B B	PR
	514226	В	PR	225	538250	B	PR
	557248		PR	2266	559251	В	HPR
	615355	C	PR	228 <b>a</b> .	637295 588250	Ç	PR PR
228b	633291	B B	PR	230a	500250	Ç	
230ъ	594254	В	PR	231	546277	<b>BBBBCTCCB</b>	R R
232	533264	В	PR	233	517250	ם פר	PR
234	487227	В	P	235	458203	D.	PR
236 <b>a</b>	425182	C	PR	2360	427184	4	PR
236 <b>o</b>		T	PR	238 2 <b>41</b>	434262	m	PR
240	415225		PR	24 <u>1</u>	479167	Ç	PR
242	442132	<u> </u>	PR	243B	432123	Č	R
243b	422113		R	2448	403097 387092	B	P
244b		. B	HPR	245			HPR
2468	344053	C	HPR	246 <b>b</b> 247b	331160		HPR
2478	334063	C	HPR	247b 248b	372096	B	HPR
2408	377102	C	PR	251a	439148		R
270	416136	T	PR	251a	447117		${\tt PR}$
るとれか	439148	<b>5</b> 5	HR	255	406081	В	HPR
256	434106		HR PR	255 257	425079		PR
2580	385070 432085	S C		258ክ	<b>4</b> 39093	T	HPR
261	528176		R P	262a	538183	В	PR
262h	562205	. 40	PR	263	584226	В	$\mathtt{PR}$
264 <b>a</b>	615256	Ca.	PR	26 <b>4</b> b		В	HR
269	555182	2 0	R	270	542168	В	PR
271	496127	T	HPR	272	465098	В	HPR
275a	508076	B	HPR	275b		C	H
276	523093		HPR	277	582136	В	PR
	************						

### Notes:

<sup>1 -</sup> Site numbers that are missing indicate that no crop was present in these sites that contained any of the species.

<sup>2 -</sup> Grid references - see table A2.1, note 1.

<sup>3 -</sup> Crops - see table A2.1, note 2. Also Bw = buckwheat

<sup>4 -</sup> Species: H = A. hybridus, P = A. powellii, R = A. retroflexus.

#### TABLE A2.3

# A KEY TO THE SYMBOLS USED TO DESCRIBE SOIL TYPES

Symbol	Name and apparent synonyms
BCL 3	Brooksten clay loam (Brookston clay)
BEL	Berrien loamy sand
BES	Berrien sand
BG	Brady gravelly loam
B <b>SL</b>	Brookston sandy loam
B <b>VS</b>	Beverley silt loam
CC	Clyde clay loam
FG	For gravelly loam (Burford gravelly loam)
GS	Granby sand
HC	Haldimand clay
HS	Huron silt loam
LL	London leam (Conover loam)
MC	Miami silty clay loam
MI	Mixed: mainly Ottawa sands and Miami silty clay loam
ML	Miami loam (Guelph loam)
PC	Perth clay (Connover clay loam)
PSL	Perth silt loam (Conover silt loam)
<b>T</b> S	Tuscola silt loam

<sup>(</sup>Based upon SOIL SURVEY REPORTS of the Canada Department of Agriculture for Elgin, Kent and Middlesex counties)

A2.2 Determination of soil particle-size distribution A2.2.1 Estimation of the specific gravity of the soil

#### a) Introduction

constants that are used to estimate the diameter of soil particles in suspension are dependent upon the temperature of the suspension and the specific gravity of the suspended soil. As a preliminary investigation the specific gravity was determined of three soil samples representative of the range of different soils that were to be analysed for particle-size distribution.

#### b) Methods

A soil sample was air-dried on a bench in the laboratory. Fifty grams of the soil were transferred to a mixing beaker and 50 ml of distilled water were added. The suspension was dispersed (without the addition of a dispersing agent) for 15 min in an electric mixer. After dispersion the soil suspension was washed with distilled water into a calibrated 500 ml pycnometer. Care was taken to keep the total volume of the suspension below about 350 ml. The suspension was heated until it boiled and maintained at boiling temperature for 10 min. At this time the heat was withdrawn and the volume of the suspension was made up to 500 ml with distilled water. The pycnometer was closed with a glass stopper and the suspension was allowed to cool to ambient temperature in a room maintained between 20° and 30°C.

When the suspension had cooled the combined weight of pycnometer and soil suspension was measured. Immediately after

weighing, the temperature of the suspension was recorded.

The pycnometer had already been calibrated and the weight of the pycnometer with 500 ml of distilled water at the same temperature was determined from the calibration curve.

#### c) Results and calculations

The results and calculations are presented in Table A2.4.

#### d) Conclusions

The specific gravities of the three soil samples ranged from 2.52 to 2.56 with a mean of 2.53. Since deviations from the mean were small the assumption was made that each soil sample had a specific gravity of 2.53.

#### A2.2.2 Sieve and hydrometer analysis

#### a) Introduction

Two parameters must be calculated in order to determine the distribution of particle sizes by the hydrometer method. These parameters are: (1) P the percentage of soil remaining in suspension at the level at which the hydrometer measures the density of the suspension; and (2) D the diameter of the smallest particles all of which will have passed the level at which the hydrometer measures the density at the time the reading is taken.

These two parameters can be determined from the following equations:

$$P = \frac{R a}{W} \times 100$$

where; <u>a</u> is a correction factor that depends upon the specific gravity of the soil and is obtained from tables,

- R is the hydrometer reading with meniscus correction applied,
- w is the weight of the soil sample.

#### D = K /(L/T)

- where;  $\underline{K}$  is a constant that depends upon the temperature of the suspension and its specific gravity. The value of K, which is obtained from tables, does not change for a series of readings that constitute a test,
  - is the distance from the surface of the suspension to the level at which the density of the suspension is being measured. The value of L varies with the reading of the hydrometer and is obtained from tables,
  - <u>T</u> is the interval of time between the beginning of sedimentation and the time at which a reading is taken.

#### b) Methods

Soil samples were air-dried in the laboratory. Fifty grams of a sample were transferred to a mixing beaker. Fifteen millilitres of 0.5N Sodium oxalate solution (a dispersing agent) were added to the beaker and enough distilled water was added to approximately half-fill the beaker. The suspension was dispersed for 15 min in an electric mixer.

TABLE A2.4

DETERMINATION OF THE SPECIFIC GRAVITY OF SOIL

	UNITS	SYMBOL			
Sample number			1	2	3
Soil type			PC	BSL	<b>F</b> G
Wt. bottle and water	g	<b>w</b> <sub>2</sub>	668.2	663.7	662.9
Wt. bottle + soil + water	g	W <sub>l</sub>	698.7	693.8	693.1
Temperature	•C	T	23.8	22.0	26.8
Wt. soil	g	Wa	50.0	50.0	50.0
Specific gravity of water		G <sub>T</sub>	0.99732	0.99780	0.99657
G <sub>™</sub> ₩ <sub>s</sub>			49.87	49.89	49.83
W <sub>1</sub> - W <sub>2</sub>			30.5	30.1	30.2
$W_a - (W_1 - W_2)$			19.5	19.8	19.8
Specific gravity of soil		G <sub>₽</sub>	2.56	2.52	2.52

$$G_{\mathbf{s}} = \frac{G_{\mathbf{T}} \, \mathbb{W}_{\mathbf{s}}}{\mathbb{W}_{\mathbf{s}} - (\mathbb{W}_{1} - \mathbb{W}_{2})}$$

Note: Abbreviations for soil types are listed in table A2.3

The suspension and any sediment were washed with distilled water into a 1000 ml sedimentation cylinder. The volume of the suspension was made up to 1000 ml with distilled water and the cylinder was sealed securely with a cork. The suspension was dispersed by turning the cylinder end-over-end for about 60 sec. The cylinder was placed upon the laboratory bench promptly after shaking and a stop-clock was started. A number 152H hydrometer was lowered carefully but quickly into the suspension in the cylinder and allowed to settle. Readings of the hydrometer were taken at the top of the meniscus after 30 sec, 1 min and 2 min.

After the reading was made at two minutes the hydrometer was removed and wiped clean. The cork was replaced in the cylinder and the suspension was again shaken for 60 sec. A second set of readings were obtained in a similar manner to the first.

The suspension was shaken a third time and set down on the laboratory bench but this time the hydrometer was not introduced immediately. At about 20 sec before two minutes had elapsed the hydrometer was lowered very carefully into the suspension. The hydrometer was brought to the expected level (based on the previous readings) before it was released.

In this way the hydrometer settled quickly without bobbing. A reading of the hydrometer was made at two minutes and the hydrometer was removed. Further readings were made at approximately the following times: 4 min, 8 min, 15 min,

30 min, 1 hr and 11:kr.

Between readings the hydrometer was placed in a 1000 ml cylinder containing 985 ml of distilled water and 15 ml of 0.5N sodium oxalate solution. At an early stage in the determination a reading was made of the hydrometer in the distilled water; this provided the "meniscus correction factor". The temperature of the soil suspension was measured also within the first 15 min of the determination.

After sedimentation was complete (11 hr), the remaining suspension was decanted and the sediment was washed through sieves of the following sizes (Canadian Standard Sieve Series): 9, 32, 65, 140, and 270. The contents of each sieve were transferred to evaporating basins and dried at 105°C for at least 12 hr. The basins were allowed to cool and the weight of each fraction was recorded.

#### Results and calculations

The results of each determination were used to construct a curve of the distribution of soil particles in the sample analysed. From the curve it was possible to estimate the percentage of particles lying within the following conventional particle-size classes:

Class	Diameters of particles within the class
Clay Silt Fine sand Coarse sand Gravel	Less than 0.002 mm 0.002 to 0.02 mm 0.02 to 0.2 mm 0.2 to 2.0 mm Greater than 2.0 mm

The description of each sample in terms of conventional particle-size classes is presented in table A2.5.

TABLE A2.5

RESULTS OF THE SOIL PARTICLE SIZE ANALYSIS

Percentage of soil (by weight) in the following particle-diameter classes (mm)

				m/ seas.		_
	1	Over	2.0 -	0.2 -	0.02-	Less than
Site No.	Soil type	2.0	0.2	0.02	0,002	0.002
1	BSL	0	l	69	24	<b>6</b>
2	BSL	0	1 2 8	70	20	8
3	BEL BSL	1	0 0 ·	<b>7</b> .9 80	8	2
4 5	BSL		9 16	73	11 8 9 7	2
<b>6</b>	BSL	1	47	45	7	<u>o</u>
7	BSL	0	24 7	49 73	21 18	<b>6</b>
1 2 3 4 5 6 7 8 9 10	BG BEL	010000	19	45 49 73 68	11	2220622222208
10	?1	ŏ	19 2 3 1 4 6	81	15 6	2
11	BEL	0	3	89 86 86	6 11	2
12	<b>?2</b> B <b>E</b> L	0	4	86	11 8	2
14	BEL	0 0 0	ć	80	10	2
15	BEL	0	6	80	12 11.	2 10
16 17	CC BSL	0	29 17	50 60	15	8
13 14 15 16 17 18	BSL	0	17 8 3	55 21	27	10
19 20	$\mathtt{BCL}$	0	3	21	43 20	33 12
20 21	BCL HC	1	36 14	31 39	20 30	17:
22	FG	0 1 0 0	20	39 68	30 8 12 7	4
22 23 24 25 26 27	B <b>SL</b>	0	45	41	12 7	4 2 2 23
2 <b>4</b>	BEL BSL	0	<b>4</b> 7	87 32	38	23
26	?2	ŏ	20	76	38 4 9 8 37 10	0
27	$\mathbf{BEL}$	0		85	9	0 2 4
28 29	FG	3 1 0	22 8	63 42	37;	12
29 30	PC FG	i	26	59	10	4 25
30 31	FG BSL	ō	5_	40	30 6	25 0
32	$\mathbf{BEL}$	0	31 7	62 34	36	
33 34	HC BCL	0	6	22	41 61	31
35	HC	Ö	2	22	6 <b>1</b>	17
36 3 <b>7</b>	BSL	0	28 22	99 21	8	4
3 <i>1</i> 38	BEL ?3	Ŏ	6	42	28	21
32 33 34 35 36 37 38 39 40 41	?3 BSL	0 0 0 0 0	4 22 8 25 37 6 2 2 2 6 1 1 8 8 1 4	34 22 22 51 66 42 77 28	12 8 28 6 7 42	23 31 17 8 4 21 4 0 21
40	BSL	0	18 8	74 28	42	21
41 42	HC BCL	0	8	13	50	29
42 43	BCL	ŏ	14	29	36	21
44	HC	0	4	19	50	27

(continued)

Percentage of soil (by weight) in the following particle-diameter classes (mm)

			CT		mm)	
	-	Over	2.0 -	0.2 -	0.02-	Less than
Cito No	Soil type	2.0	0.2	0.02	0.002	0.002
Site No.	DOTT ALDA					
		•	•	AE	2.4	19
45	HC	0	2	45	34	19
46	HC	2	58	26	10	4
A7	HC	2	33	30	14	23
46 47 48	BSL	Š	7	30 65	18	8
40	77.T	2 0	Ė	<b>1</b> 9	30	23 8 37
<b>4</b> 9	HC	Ū	2	19	14 18 39 58	20
50	HC	Ü	Ŧ	12 46	20	29
51	BCL	0	5	46	35	14
52	?4	0 0 0 0 0	2 58 33 7 5 1 5	31	35 <b>41</b>	<b>27</b>
52	BEL	0	33	49	11	7
)) 54	BEL	Ŏ	33 46	46	6	2
2 <del>4</del>	DOT	ĕ	16	42	20	1 <b>7</b>
55	BSL	0	72	46	Ā	<u> </u>
56	?3	Ō	15 26 16	00	Ž	7
5 <b>7</b>	BEL	0	16	76	0	<b>6</b>
49 50 51 52 53 54 55 56 57 58 59 60 61	?1	0	23 8	60	9	0.
59	BEL	ı	8	28	33	31
60	?1	Ō	30	61.	7	2
61		_0	ำำ	77	8	2
OT.	BEL	2	<u> </u>	86	ž	2
62	BEL	<u>U</u>	3	31 49 46 42 66 76 60 28 61 77 86 85	11 6 20 4 6 9 33 7 8 3	2
63 64 65 66	BEL	00010,2000000	30 11 9 3 9	05	10	72174283122222662862824220
64	$\mathtt{BEL}$	0	9	74	10 10 2 24	6
65	BSL	0	10	74	TO	0
66	BEL	0	7 2 16	89	2	2
67	92	Ŏ	2	70 66	24	8
CQ	?2 ?2	Ŏ	ī6	66	12	6
68 69	1.2	0	11	23	41	23
09	?3	0	77	<b>57</b>	<u>16</u>	8
70	CC	0 1 1 2 0	19 51	21	7	õ
71	BSL	0	.5 <b>1</b>	42	7 19 12 18 5 27	<u> </u>
72 73	HC	1	23	50	73	4
73	BEL	1.	19	66	<u> 15</u>	2
74	BEL	2	15	<b>6</b> 3	18	2
75		7	19 15 69 22	50 66 63 26 <b>2</b> 4	5	0
12	BG	0	22	24	27	27
(0	HC	0	22		19	6
$T_{1}^{T}$	BSL	U	2)	61	26	6
78	BSL	0	7_	67 01		Ŏ
<b>79</b> )	BEL BSL	0	17	10	2	Ö
80	BSL	0	27	68	3	č
81	BEL	O.	6	78	10	0
82	DQT.	Ť	5	83	11	Ō
82	B <b>SL</b>	<b>☆</b>	์ า	8ō	7	2
<b>V</b> 3	BEL	Q	<b>⊘</b>	90	8	0
04	BEL BEL	0	<u>.</u>	50 61 78 68 78 80 90 84 69 87	10	0
05 05	BEL	Ŏ	ō	24	<u> </u>	2
86	BEK	0	<u>9</u>	60	20	<u> 7</u>
87	$\mathtt{BEL}$	0	7/	69	20	7
74 75 76 77 78 79 80 81 82 83 84 85 86 87 88	BEL	0000100000	23 7 17 27 6 5 11 26 9 7 9	87	19 26 5 3 10 11 7 8 10 5 20 2 38	6 6 0 2 0 0 2 0 2 4 <b>2</b> 2 2 2
89	HC	Ŏ	. 16	24	38	22
<del>4</del> 5	ЩО	•		•		

Note: 1 - A key to the abbreviations used to describe soil types follows table A2.1

TABLE A2.6

# ADDITIONAL RECORDS OF THE OCCURRENCE OF A. HYBRIDUS The following observations, when combined with those

in tables A2.1 and A2.2, complete the records of occurrence of this species as determined in this study.

Grid reference*	Habitat.	Grid reference*	Habitat
<del>3</del> 51303	Garden	356187	Garden
437146	Corn field	458183	Garden
506189	Garden	525186	Bean field
587249	Bean field	711410	Bean field
807677	Farmyard	853415	Bean field
*Note:			

For explanation of the grid reference see table A2.1, note 1.

#### APPENDIX 3

#### GERMINATION DATA

The following tables contain the results of germination trials conducted in experiments 1 to 13. In most cases the results are presented as cumulative daily (or weekly etc.) totals of the number of seeds germinated in each replicate. Where an estimate was made of the viability of ungerminated seeds at the conclusion of a trial, this figure has been combined with the final germination total to provide a value for total viability. In those trials in which viability was not estimated, the number of seeds counted into each replicate has been recorded.

#### Abbreviations

Abbreviations have been used to describe the levels of the various factors examined in many of the experiments. The following is a list of these abbreviations with their explanation or the page number on which they are explained in the text:

Collections: B1, B6, H7, P1, P2, P3, P4, P5, P6, R1, R2, R3, R4, R5 and T1.

These identify the species and the source of the original material and are fully described on page 208, in table 6.1.

## Light regimes (experiments 6 and 10)

- LD or L/L daily alternating light/darkness cycle maintained throughout trial.
  - L/D Daily alternating cycle maintained for seven days then transfer to continuous darkness.
- D or D/D Continuous darkness throughout the trial.
  - D/L Continuous darkness for seven days then transfer to alternating light/darkness.

## Wintering treatments (experiments 9 and 13)

- B or -15 Seeds buried 15 cm beneath soil surface.
  - -60 Seeds buried 60 cm beneath soil surface.
- S or OO Seeds placed on the soil surface.
  - +15 Seeds suspended 15 cm above soil.
  - +60 Seeds suspended 60cm above soil.
    - P Seeds left on remains of parent plant.
    - L Seeds stored in the seed-storage room.
    - +R Seeds that wintered in the presence of remains of the inflorescence.
    - -R Seeds that wintered in the absence of remains of the inflorescence.
    - BB Seeds buried for two winters and the intervening summer at 15 cm below the soil.
    - SB Seeds placed on the surface for first winter then buried for the following summer and second winter.

# Position of seed (experiment 10)

- T Seeds taken from the terminal inflorescence.
- B Seeds taken from a single basal, lateral inflorescence.

# Retention of utricle (experiment 10)

- + Germination of seeds with utricle retained.
- Germination of naked seeds.

# Incubator shelf (experiment 9)

- U The upper of two shelves.
- L The lower of two shelves.

#### Temperature regimes

- 35/20 A day temperature of 35°C and a night temperature of 20°C.
- 30/15 A day temperature of 30°C and a night temperature of 15°C.

#### General abbreviations

rep. replicate.

A. tuberculatus T1 Rep.

A. retroflexus R1 Rep.

A. powellii P1 Rep. 4

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2 - TN = Mumber of seeds used.

3 - Other abbreviations are listed on page 516.

# TABLE A5.1

A. retroflexus A. tuberculatus RA Rep. Rep. CUMULATIVE GERMINATION SCORES OF SEEDS OF EACH COLLECTION IN EXPERIMENT 1 Rep. A. powellii P1 Rep. 4 A. blitoides
BREP.
1 2 3 4 A. blitoides B6 Rep. Day<sup>1</sup> 1

CUMULATIVE GERMINATION SCORES OF SEEDS OF EACH COLLECTION IN EXPERIMENT 2 TABLE A5.2

Days from the beginning of the experiment (Day 0 = 5th October, 1966)

	100	100	100	100	
7 0 0 10 11 12 13 14 15 16 18 19 20 21 22 23 26 34 40 47 56 71	7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7	1 0 0 2 4 5 6 6 7 77 77 78 81 81 82 82 82 82 04 71 77 45 45 46 10 2 0 0 1 3 16 32 34 34 77 77 78 81 81 82 82 82 82 84 54 77 77 78 70 10 10 10 10 10 10 10 10 10 10 10 10 10	00 1 2 2 2 2 2 3 6 7 9 11 12 10 2 1 2 2 2 2 2 2 2 6 10 0 0 0 1 2 2 2 2 2 2 2 5 5 5 6 6	0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	5 5 5 5 5 5 5 6 6 6 5 5 2 5 2 4 4 4 4 4 4 7 7 7
		R1		R2	

Note: Abbreviations are listed on page 510.

TABLE A5.3

3 CUMULATIVE GERMINATION SCORES OF SEEDS OF EACH CODLECTION IN EXPERIMENT

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	103	52 52 52 52	47 71 72 64	70 92 92 96	223	7,400 4,680 4,680	56.00
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	2	55 52 52	5865	70 66 72 66	2027 2027	4404 7000	565 655 1
	8	54 54 57 57 57	8093	929 929 967 967 967	2022	222 2027	500 500 1
	59	4 2 2 2 2 3 3 4	2267	5828 8288	2022	2967	800 100 100 100
	49	55 55 52	2472	2828	2232	<b>8</b> 4 <i>万</i> 0	860 800 100 100 100 100 100 100 100 100 10
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( pag sted 17:5 Abbrevictions

Note

CUMULATIVE GERMINATION SCORES OF SEEDS OF EACH CODLECTION IN EXPERIMENT 3

TABLE A5.3

TABLE A3.4

# CUMULATIVE GERMINATION SCORES OF SEEDS OF EACH TREATMENT IN EXPERIMENT 4

												No. of		
Treatment	Rep.	1_	2	3	4	5	7	9	11	14	18	22	25	
Black seeds	1 2 3 4	1 2 1	13 13 10 9	14 13 10 9	17 18 14 13	24 24 25 23	24 24 25 23	24 25	24 25	24 24 25 25	24 25 25 25	24 25 25 25	25 25 25 25	25 25 25 25
Brown seeds	1 2 3 4	0 0 0	0 0 0	0 0 0	0 0 0 0	0 0 0	0 0 0	0 0 0	0 0 1 0	0 0 1 0	0 0 1 0	0 0 1 0	0 0 1 0	1 0 1 0
Small seeds	1 2 3 4	0 0 0 0	4 8 2 6	<b>4</b> 8 2 6	9 12 5 10	23 21 20 24	21 20	22 20	22 20	23 20	24 22	24 23	24 23	25 24
Insect- eaten seeds	1 2 3 4	0 0 0	0 0 0	0 0 0	0 0 0	0 0 0	0 0 0	0 1 0 0	0 1 0 0	0 1 0 0	0 1 0 0	0 1 0 0	0 1 0 0	0 1 0 0

Note: Abbreviations are listed on page 10.

TABLE A3.5

TOTAL GERMINATION SCORES OF SEEDS
OF EACH COLLECTION IN EXPERIMENT 5

Collection	R R	epli 2	cate 3	4	
P2	74	73	65	5 <b>7</b>	
P3	42	49	43	41	Total seed
Rl	91	96	96	96	used = 100
R5	67	74	84	85	

TABLE A5.6

CUMULATIVE GERMINATION SCORES OF SEEDS OF EACH TREATMENT IN EXPERIMENT 6

Days from the beginning of the experiment

	Number of seeds used	0000	9000	7,700 1,000 1,717	7777 0000 0000	7770 7000 7000	0000	7777 0000 0000
	47	25 24 26 26 26 26	788 188 188	0000 0000 0000	9939 9329 94	98 97 76 76	72 72 83 84	0 0 0 0 0 0
	32	3473	7 7 7 8 8 8 9	9995	0000 0004	98 92 92 92	72 77 83 84	77.00 64.00 70.00
	28	2473 2473 2473		999 999 999	0000 0004	92 92 92	72 77 83 84	7 7 7 8 9 9 9
1967)	23	2473 2473 2643	7 2 2 3 8 7 8 8	999 999 999	0000 0004	988 988 7	72 77 83 84	
	20	2473	7 7 7 8 8 9 8 9 8	9999 9997 9697	0000 0004	9899 8899	72 73 83 83	47.00 47.40 7.40
January,	12	25 173 26	7 7 8 7 8 7 8 7	999 969 97	9000 0000 4	887 82 82	24 88 88 88	ろうる ろうはら
	80	24 24 25 25 25	477 477 8	999 999 7	9000 9004	52828 52828	69 77 77	ろり ろり ろり り
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II O	9	2524		999 999 95		44 45 450 484 484 484 484		7.000 7.000
Day	7	5°5%		999		4444 44704		50 47 60 60
O	77	4 26 33 33		90 90 90 90 90 90		300 800 300 800 300 800		48 573 573 58
	2	20 20 20 20 20 20 20 20 20 20 20 20 20 2		9000 4000 7000		2288		44 72 72 72 72
	~	87.000		000N		9777		φ00C
	Rep.	-0W4	− 0 m 4	<b>∠</b> 01√4	← 01 m 4	<b>-</b> 0w4	<b>ころり</b>	7 N N 7
	Light	1/1	L/D	D/D	D/L	L/L	T/D	D/D
	Collection	P2				F 23		

ÕŌ	9999	9000	9999	7777	7770	9000	9999	9999	7777	7777	700
96 95	9999 922 4	998 972 44	72 77 83 84	557 66 66	786 100 100	9898 925 935	87 92 86 86	62 62 62 62 63 63 63 63 63 63 63 63 63 63 63 63 63	0000 0000 00000	900 907 94	CO
96 95	9999 922 4	92738	72 77 83 84	557 64 60 64	988 922 100	8080 9000 9000	89 92 86	625 625 625 625 625 625	9999 9989	90 90 90 80 80 80	Q
96 95	0000 0000 4	989	72 77 84 84	2 2 2 4 6	87 91 98 100	89 95 93 93	87 89 86 86	6285 6285 6285	9999 9889 9	9000 4000 2000 4000	CO
		95 98 92 92			0			.0.010101	- 01001	M+M0	C
96 95	9626	9899 9833 9833	72 77 83 83	57.00 6.75 7.00 7.00	88 97 97	983 93 93	869 867 867	622 622 622 623 623 623 623 623 623 623	2888 2888	2288 888	C
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	D/T		L/D						D/L	L/L	•
		P 3				R1				R5	

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D O O	9999	9999	9999	7,700	7000	7,700	7779	7777
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92 98 100	98989 97777	86 92 86 86	65 65 65 65 65 65	0000 0000 0000	996 986 93	92 98 98 98	58 57 57 58	978 989 899
97 98 100	89 95 93	87 92 86 86	62 62 62 62 63 63 63 63 63 63 63 63 63 63 63 63 63	9 9 9 9 8 8 8 8	9000 4000 7000 7000	98 98 98 98	82 82 72 73	978 989 899
96 98 100	87 93 92 92	87 92 86 86	622 622 622 622 622 623 623 623 623 623	94 96 98 98	0000 0000 0000	988	85 87 77 77	78 91 89 89
88 97 97	893 92 92 92	87 92 86 86	622 622 623	9999 2007	9000 74800	98 98 98	82 82 72 72	97 98 80 80 80
86 96 94	880 927 927	86 92 86 86	62 62 62 62 63	9999 7999 7999	0000 00000	98 98 98	52 52 53 54 54 54 54 54 54 54 54 54 54 54 54 54	77 90 97 87
49 64 63	25 25 25 27 27	86 92 86 86	625 625 627 627	855 87 87	90008 0000	90799 90708	827.7	00 00 00 00 00 00 00 00 00 00 00 00 00
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ne ( ce	L/L	L/D	מ/מ	D/L	L/L	L/D	מ/מ	D/L
	R7				<b>ሕ</b> 5			

Note: Abbreviations are listed on page 510.

TABLE A3.7

CUMULATIVE GERMINATION SCORES OF SEEDS
OF EACH TREATMENT IN EXPERIMENT 7

Wintering conditions	Rep.	1	<b></b> ∶2	3 3	ays <sup>1</sup>	5	6	7	No. of seeds used
Laboratory	1 2 3 4 5	0 0 0 0 0	1 2 1 0 0 7	4 3 3 1 3 8	7 4 5 2 4 9	7 5 5 2 5 9	8 5 5 7 9	9 8 5 4 8 10	50 50 50 50 50 50
Field	1 2 3 4 5 6	0 0 0 0 0	14 11 8 18 25 12	21 15 23 25 31 18	24 16 23 26 32 21	24 17 23 26 32 22	24 18 23 27 33 22	25 18 24 27 33 22	50 50 50 50 50 50

Notes: 1 - Days from the beginning of the experiment, day 0 = 25th May, 1967.

<sup>2 -</sup> Other abbreviations are listed on page 510.

TABLE A5.8

CUMULATIVE GERMINATION SCORES OF SEEDS OF EACH TREATMENT IN EXPERIMENT 8

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experiment	7	242							
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	12	4 200 200 200 200 200 200 200 200 200 20							
the	7	450						000	
of	19	222				<b>5</b> 83		200 200 200 200 200 200 200 200 200 200	
beginning	6	38%				47 48 47		\$ 65 \$ 54	
inn	ω	28 28 25	525			45 45 46		5 5 5 5 5 5	00 m
oeg:	2	27 28 22	52 54 55 54	344	<b>表</b> 8 表	543		-	
the 1	ဖ	966		2 \$ \$	383			42 47 47 5	49 48 5 47
	r/	242	722	22 25 25	525		222	27 46 46 54	2 49 2 47 5 46
${ t from}$	4	ဖ၀၀	744 747	804	<b>752</b>		555	67 4 7 7 7	
	3	NO0	3%5	007	28 47 45	272		500	454
Days	N	000	N00 N	000	222	N0/2	29 22 22	41/0	13 17 20
	~	000	770	000	004	700	0 mc	000	070
	Rep.	<b>←</b> Ωκ	1 - Un	7 700	-0m	1 - WW	7 UM	7 N N	70M
	Temperature (°C)	25	35	25	35	25	35	25	35
	0.00 to 0.00 t	i		8-day dry		15-2-67 4-week wet storage	)	7 4-week dry storage	i
Date of	_	18-1-67		26-1-67		15-2-6		15-2-67	

TABLE A5.9

MENT IN EXPERIMENT 9, TRIAL 1	eginning of the trial	0 = 27 <sup>th</sup> Oct., 1967)	5 14 15 16 17 10 12 50 51 51 51	7 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	007				2 2 2 2 2 3 3 3 2 2 2 2 2 2 2 2 2 2 2 2	7 1 1 1 7 3 3 3 3 6 6 6 6 6 6 6 6 6 6 6 6 6 6 6	2 2 2 3 3 3 4 4 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1
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Note: Abbreviations are listed on page 510.

Z Collection

		ł									
Ö	Number of viable seeds	<b>353</b>	, <u>r</u> rrr	2007 100 100	885	<b>\$</b> 50	200 101 101	24 g 27 g 27 g	987 107 104	222	56
TRIAL	d	\$ 5°5	, <b>Ľ</b> 88	8 8 8 8 8 8	<b>353</b>	\$\$C	8 8 8 8	348	88 96 96	222	<b>3</b> 5
•		523	22Z	888	<b>353</b>	<b>35</b> 8	83 94 97	<b>44</b> 6	8860	222	<del>2</del> 6
6 IN		522	7. 2.2.2	74 78 85	<b>252</b>	<b>35</b> 8	<b>8</b> 48 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8	<b>448</b>	83 85 85	222	\$ C
RIME		5 253	222	\$25 837 837	<b>575</b>	<b>3</b> 50	83 84 92	245	82 85 85	888 8	4 2 5
experiment		7 5 5 3 2 5 5 3	. <u>۲</u> 88	282	<b>353</b>	54°C	82 98 94	248	82 78 85	222	46 50
H	<b>0</b>	4 2°C 3	. <i>2</i> 88	63 77	<b>222</b>	444 49 49	88 88	2889	79 77 82	<b>722</b>	46 50
MENT	4 <del>7</del> 7	I	. <i>2</i> 88				•		333		
TREATMENT	_							2389	232	522	46 C
	nn P							34 17 17	73	22%	45 C
F EACH	e II	1	5 <b>23</b> 2					288	689 72	200	37 50
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3 0 F	Days		ν σπο								
SCORES	н	1	4 4 6 6 6 6 6 6 6 6 6 6 6 6 6 6 6 6 6 6							80°C	26
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GERMINATION			5 4 2 Kr								1
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Mote: Abbreviations are listed on page Lu.

TABLE A5.11

CUMULATIVE GERMINATION SCORES OF SEEDS OF EACH TREATMENT IN EXPERIMENT 9, TRIAL 3

Days from the beginning of the trial

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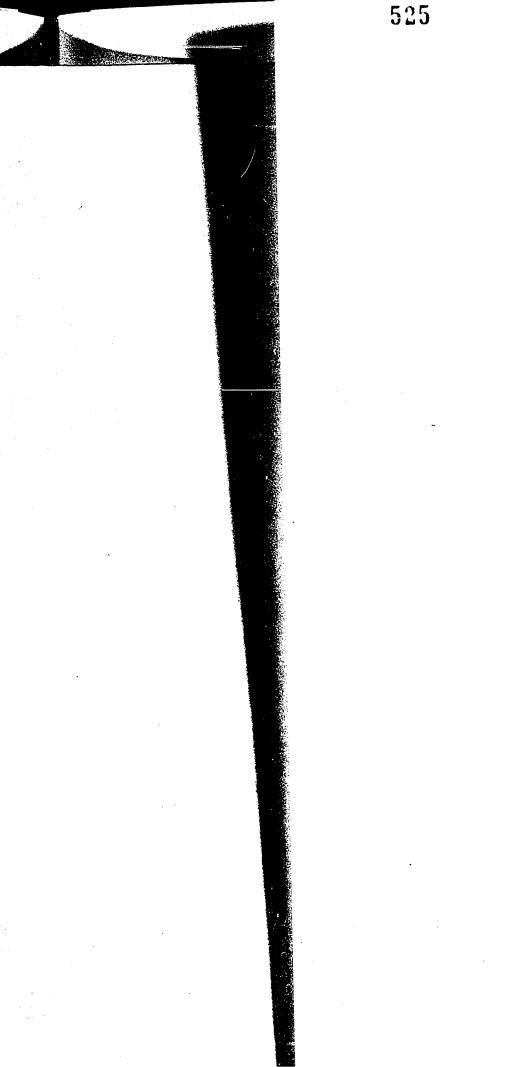
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Note: Abbreviations are listed on page 510.

TABLE A5.14

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SCORES			7	27	0	00	0	00	00	0	00	00	<b>&gt;</b>	000	0	00	00	0	00	0	001
CHMULATIVE GERMINATION			,	Replicate	<b>~</b>	OI M	7	~ (	Mν	<b>.</b> ;	· <b>-</b> 0	1 <i>w</i> -	<b>4</b> <i>7</i>	- O K	/4	<i>-</i> ∼ <i>N</i>	<b>M</b> =	t <	Иĸ	/4	<i>−</i> 011
CIMULATIVE				Collection	2	I		P2			P3			ታላ ተ		P5		94	)		R.

3822 0448 6494 8429 8424 8346 8414810 2877 GOLO 0450 O485 O551 8550 GO50 FO018510 8777 7089 7708 8707 77770 01771000 8909 70012710 2864 2664 277 8487 3×2× 8×2× 10+4 401/ 401/ 12/60 17/00 ×20012/10 3485 8888 500- 600F 8005 5050 008+ 800005-0 3888 8988 6594 6984 9500 6978 9558 9000000 4040 MUNU NTUO CUMB 0407 MUBT MUNU UU000000 WWUO ORUO OOOO OOUU OOOO LOOO OOOO LUOOOOOO **トロアナ トロアナ トロアナ トロアナ** H7 R5

**P6** 

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TABLE A3.15

CUMULATIVE GERMINATION SCORES OF SEEDS OF EACH TREATMENT IN EXPERIMENT 11

the experiment	1968)
om the beginning of	1 - 15th Way
Days from the	

0	seeds used	7000 7000 7000	500	WWW.	7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7	200		77777 00000 00000	20000 20000 20000	20000 20000 20000	
	88	242 424 424	12	123 173	1 on5	- ω	0000	112 103 103 604	79 75 74 67	4404 7404	О п∕ ≤
	83	25 45 45 45	15	1787 1787	1 on5	- ω	0000	112 109 96 57	23 23 53 53	5584 584	o <i>∿</i> -
	25	545	15	207 207 1	1 ont		0000	112 109 94 56	33 33 66 66	2484 0784	04:
	68	545	5	2017 r	/ ® ~ £	. 0	0000	112 109 93 56	22 23 66 66	827 67 70 67	04:
968)	61	545	7	207 207	+ ω 4 <u>5</u>	- rV	0000	111 109 93 47	522 62 62 62	800° 800°	0 W =
<b>د</b>	53	802 827	7	200 200 200 200 200 200 200 200 200 200	t 040	2	0000	252 4675 4675	0 0 0 0 0	2020 2000 2000	00=
ր ⊯ay	46	2004 2007	7	25-6-	4 4 4 4	Z.O.	0000	<b>8</b> 273	5000 5000 5000	1201 2014	000
15th	44	08 80 28 80	16	9440	0 0 0	0	0000	55 48 88 48 84	700°C	72 27 8 27 8 27	000
II O	38	18 20	19	4 5 6 6	000	00	0000	4 4 8 8 8 8 8 8 8	258 258 24 26 26 26 26 26 26 26 26 26 26 26 26 26	9729	000
(day	55	W40	30	w	000	00	0000	28 44 22 22 22	28774 8774	770M	000
_	28	W40	30	w	000	00	0000	2448	4444	770M	000
	50	700	- ر	0400	000	00	0000	2524 2021 8	2234	7707	000
	16	000	00	0000	000	00	0000	5276	0007	0000	000
	9	000	00	0000	000	00	0000	8773	0000	0000	001
	Rep.	<b>-0</b> 4	<b>0</b> 4	L 01 W :	4 701	<b>0</b> 4	<b>-</b> 0万4	−0v4	<b>-004</b>	<b>と</b> るる4	<b>∠</b> 01
	Soil Collection	P3		표	Т?		Control	P3	<b>R</b>	H7	Control
	Soil	BES						BEL			

500 500 500 500		20000 00000	20000	00000 00000		00000 00000	0000 0000 0000	2000 2000 2000 2000
44 29 45 59	o <i>∿</i> 4∠	86573	¥355	ωω <del>77</del> ωω	4007	75 48 72 74	25 25 25 25 25 25 25 25 25 25 25 25 25 2	0228
45 28 44 44	o <i>∿</i> 4∠	25.88	79% 70% 70%	8 <sup>7</sup> 1 <sup>3</sup> 8	W00 C	4447 4607	240 240 240 240	00000
45 45 43 43 43	0447	2248	1932	2 <sup>7</sup> 2 <sup>7</sup> 8	000 <del>-</del>	か447 4どのご	75 76 70 70 70	00/200
28 27 27 40	0447	25 25 26 26 26	2000 2000 1000 1000 1000 1000 1000 1000	67178	0005	4444 4667	75 74 80 80	00000
3 3 3 3 3 3 3 3 3	0 1 4 4	25 25 25 25	1888 15	245	000	2444 2464	252 20 20 20	0000 0000
222 223 223 223	0 <i>0</i> 47	74 47 56 23	28 40 15 15	299	7007	2884 2884 20	777 727	0 14 25 25
123 123 14	0007	77 47 28	9632 520	2000	7007	08846 0870	128 178 179	0 4 4 8 9 8 9 8 9 8 9 8 9 8 9 8 9 8 9 8 9
17 18 13 13	0000	26834 2004 2004	087N	らてろろ	0007	2007 1000 1000	0477 427	0 maa
9 97 9								
770M								
770M								
0-	0000	0860	0000	0000	0000	L48L	0000	0000
0000		0070						
0000								
·								
Н?	Control	P3	R 7	2н	Control	P3	ጸ <b>ኅ</b>	Н?

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TABLE A5.15 (continued)

the experiment 1968)
from the beginning of (day 0 = 15th May,
Days fr

Number of seeds used				WWW 0000	500	2000 2000 2000	200	, 200 200 200 200 200 200 200 200 200 20	•			WWW 0000	200	2000 2000 2000	500 500
M 68	l	200	N	2228	0	らて2	α·	400	- (	000	) M	1800 1800 1800 1800 1800 1800 1800 1800	38	74 24 24 24	23
83	1													\$255 \$200	
ر بر														3283	
α	1													3223	
7														22.0%	
א ע														9448	
, g	1	000												ο <u>ν</u> 4%	
`	1	000	N	μμοα	) 0	070	0	000	)	00	00	0 14 43 43	3	ดอดอ	77
0	۶ R													0.00	
1	3													0000	
,	2	000	0	7007	- c	000	0	000	)	00	00	000	12	ONOR	00
(	2	000	0	0000	<b>.</b>	000	0	000	)	00	00	700	2 ω	0700	00
•	16	000	0	0000	<b>.</b>	000	) C	000	0	00	00	000	0	0000	00
1	0	000	00	0000	<b>5</b> C	000	) C	000	0	00	00	000	0	0000	00
	Rep.	<b>-</b> 04	<b>0</b> 4	- 40W	<b>4                                    </b>	- 0 m=	t <	- 010	4	<b>∠</b> 0	W4	- <b>C</b> UK	<b>ク</b> 4	- てころか	+ <- (
	Collection			P3	ţ	<b>ਸ</b>	60	<b>,</b>		Control		P3		ᅜ	H7
	Soil			HC								PSL			

							i
500	0000 0000 0000	0000 0000 0000		0000 0000 0000	2000	0000 0000 0000	
98 38	22 24 24	03470 100	ดอพด	. 136 98 98	2004 2004 2004 2004	W77W 7	-477
96 36	25 27 24 45	8446	nona	87 134 98	いい40 いい40	0.73 0.78 0.78 0.78 0.78	-466
96 36	25 24 24 24	0 0 0 0	nona	74 133 96 96	ひい4 @ 4 ぴぴぴ	2748 277 277	-4
96 36	25 29 29	4400	nona	66 133 97 73	でで40 い404	272	-4
64 35	2272	4400	ดอพด	6 6 6 7 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8	でで4.0 とどの4	24 74 74 74 74	-4
60 35	24 26 28	0000	00m0	64 64 58	44 W4 7/2 08	7728	0777
ÖÑ	.~2. + %	w w O 70	010WZ	55.23 20.23 20.23	445 445 484 484	92728	0777
43 29	ดอดอ	770M	-0	52 20 45 62 63 63	800 800 900 900	7227	7000
26 26	0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	070N	7070	2004 2007 2007 4	2002 2002 2002	<b>~</b> 0₩0	0000
23	0000	0000	0000	2844	あるなって	0070	0000
20 12	0000	0000	0000	247 2643 200 200 200	2000 2000	0000	0000
						0000	
						0000	
000	0000	0000	0000	0 - WC	0000	0000	0000
						+ -UW4	
	ᅜ	Н7	Control	P.	ፑ	Т.	Control

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TABLE A3.16

CUMULATIVE GERMINATION SCORES OF SEEDS OF EACH COLLECTION IN EXPERIMENT 12, TRIAL 1

g.33-ation	Plant	1	25 <b>-</b> 9 Re	-68 p. 3	3 4	23 1	Rer 2		3 4	Number of seeds sown in each rep.
Collection	1	0	0	0	0	0	0	0	0	100
<b>P1</b>	2	000	000	000	0	0	0	0	0	100 100
		0	0	0	0	0	0	0	0	100
P2	1 2 3	000	000	000	000	0	0	0	0	100 100
•					0	0	0	0	0	100
P3	1 2 3	000	0 0 0	0 0 0	000	000	000	000	0	100 100
<b></b> .		0	0	0	0	0	0	0	0	100
<b>P</b> 4	1 2 3	000	000	1 0	000	0	0	1	0	100 100
			0	0	0	0	0	0	0	100
P5	1 2	0	Ö	Ö	ŏ	Ō	0	0	0	100
<b>P</b> 6	1	0	0	0	0	0	0	0	0	100 100
	2 3	0	0	0	0	0	0	0	0	100
774	2 1	0	0	0	0	2	12	2 0	0	100
R1	2	0	000	0	0	2 0 0	2 4	· 4	1 4	100 100
7.		0	0	0	0	0	0	0	1	100
R4	1 2	0	0	Ö	ŏ	Ō	0	0	0	100
R5	. 1	0	0	0		0	1 2	4	0	100 100
	2 3	0		0		0		Ö		100
ττΩ	2 1	0			_	0	0			100 100
Н7	2	0	0	0	0	0	0			100

Note: Abbreviations are listed on page 510.

TABLE A3.17

CUMULATIVE GERMINATION SCORES OF SEEDS OF EACH COLLECTION IN EXPERIMENT 12, TRIAL 2 Days from beginning of trial (Day 0 = 18th Septermber, 1968)

Number of viable seeds	<u>5</u>	/;4 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.	+	ගි දි	1 4 4 0 0 8	) [	7.4 7.7.	50 50	48	48 48 48	53	<del>5</del> 2	- 60	46	40 00 00	2,4	64	91	O.C.	×4 8	50
10	12	<del>2</del> 25	<u>,</u>	70	- 0 k	2	ο£8	54.5	10	ろろ	56	07	- N	~	3	7 C	- W	0	Оп	701	~(
6	12	223	4	16	ころっ	3	968	545	10	- ww - cw	26	07	- N	<b>~</b>	3	7 C	- W	0	04	<b>⊦</b> ⊘	~ (
· <b>ω</b>	7	27.	7	16	C C v	2	96	252	70	- でと	25	01	- N	~	100	07	- <-	0	0 =	t (2)	<b>~</b> (
6	7	22	7	16	20 20 20 20	2	96	22 23 23	7	220	24	0	<del>-</del> α	~	3	07	- ~	0	0 =	t 01	~
G	7	228	13	16	2.0 2.0 2.0	ξ,	17	200	7	- 20 で 00 00 00 00	24	0	<b>~</b> α	<b>~</b>	0	0 7	- ~	0	0 =	t <<	<u></u>
ហ	0	,25	7	73	65,	72	12	<u>66</u>	, (	788	72	0	<del>ر-</del>	<b> </b>	N	0	) C	0	0 K	na n	~
77	- σ	, <del>, , , , , , , , , , , , , , , , , , ,</del>	2	12	94,	<del>ر</del>	70	77	` (	270	17	0	00	, <del>C</del>	~	0	0 ~	0	07	- 0	<b>~</b>
, к		~ 0	و.	2	96.	ဖ	40	. დ დ	) (	Nοα	0 [	. 0	00	0	~	0	00	0	00	00	0
C	u с	000	0	0	07	0	0 ~	07	-	000	0	0	00	0	0	0	00	0	00	00	0
7	-		0	С	00	0	00	000	>	000	00	0	00	) C	0	0	00	0	00	00	0
ſ	Kep.	- 01 K	/4	~ ~	- U M	4	<u></u> 0	1 w =	‡	<b>∠</b> 01	<b>~</b> 4	٠ ٣	01	クユ	~	· 0	<i>ω</i> =	t   <	40	<b>04</b>	~
	Plant	<del>, -</del>		C	J		77			4		7			n	J		7	`		4
	Collection	Ā										ф С	J +								

/v4 vv44 4vvv v444 vvvv vvv4 4v44 44v4 4444 vvv4 4: >>> vv000 4vvv v444 vvvv vvv4 4v44 44v4 4444 vvv4 4: し からろし からろし からろし からろし からろし からろし からろし かんご S 3 S 4

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Days from beginning of trial (Day 0 = 18th September, 1968)

(Continued)

TABLE A5.17

Note: Abbreviations are listed on page 510.

Number of viable seeds	47	4 4 6 5 0 0 0	47	-64 -62 -63	44 54 50 54	48 47 47 47	44 42 47 47	444 7460 760	449 50 49	48 48 53	4 4 2 4 4 2 4 2 4 2 4 2 4 2 4 2 4 2 4 2
10	5,	244 びんこ	42	282	4 4 4 5 5 4 7 4 7 4 7 4 7 4 7 4 7 4 7 4	2882 2987	8233	2002	24 24 24 26 26	233 247 268	22 22 23 23 24 24
6	27	22 25 25	33	£22	445 445 445 445 445	2258	2243	22 28 28 28 28 28	44 44 47 47 847 48	23377	200 200 200 200 200 200 200 200 200 200
ω	19	75 26 71	22	217 255	8248	25 20 13	<b>2444</b>	9526 1984	45 45 45 48	23377 25377	2862
2	14	9 26 26	29	2,8 2,4 3,6	7222	7772	24 24 74 90	5565	3473	16 23 24 24	2775 2000
Q	7	7 9 7	7	646	8777	. <u> </u>	22 22 24 26 27	תאשת	2824 2824 294	26 20 40 40 40 40	√4 √∞
ហ	0	0 W ()	l <b>~</b>	ωα <i>-</i>	W40k	1 C 4 WC	440C	000	22 15 24 9	75 73 73 73 73	7700
7	+ 0	007	. 0	₩ C	, 7000	-001	<b>.</b>	0000	∞ <i>w</i> ∞∠	977	0000
и	70	7000	- 0	070	0000	0000	0770	0000	0000	00°C	0000
c	V С	007	- 0	000	0000	0000	000	0000	00-0	0000	0000
<	- c	000	o 0	000	0000	0000	0000	0000	0000	0000	0000
í	Rep.	- N M -	4 ~	· U M =	4 -uw=	† -aw=	t -0w4	- <b>-</b> 0 <b>m</b> 4	- てころせ	. ていひみ	− 0 m 4
	Plant	Ŋ	К	`	~	α	~	a	М	<b>#</b>	~
	Collection	7.H	.*		R4		R5				<b>L</b> H

TABLE A3.18 CHMULATIVE GERMINATION SCORES OF SEEDS OF RACH TREATMENT IN EXPERIMENT 12, TRIAL 3

	Temper-										
Collection	ature 1 regime	Rep.	( as	3	4	5	6	7	88	15 <sup>1</sup>	seeds
Pl	35/20	1 2 3	0 0	6 10 9	14 16 15	17 18 18	19 21 18	20 26 20	20 27 22		47 45 47
	30/15	1 2 3	0 0	000	1 0 1	1 0 2	1 2	1 2	1 1 2	18 3 23	48 <b>49</b> <b>4</b> 9
P2	35/20	1 2 3	0 0 0	0 0	0 0 1	0	0	0 2 1	0 2 1		49 47 45
	30/15	1 2 3	0	0	0	000	0	0	010	2 4 7	47 50 49
<b>P</b> 3	35/20	1 2 3	0 0 0	000	0	0 0	2 4 3	2 4 4	2 4 7		46 47 48
	30/15	1 2 3	0	0	000	0	0	0	0	7 7 5	47 47 46
R1.	35/20	1 2 3	0	010	4 2 1	8 9 3	13 17 8	3 22 4 18 20	31 28 30	; )	42 39 46
	30/15	1 2 3	0 0 0	000	0		5 3 0	6 4 0	6 7 0	39 32 46	42 33 49
н7	35/20	1 2 3	0	. 1	6 3	8 4 0 4 1	4 4	4 44 8 2:	4 40	S L	50 50 48
	30/15	1 2 3	0	0 0	. 2	2 4	. 4	. 4	2 4 1	37 38 18	47 50 48

Notes: 1 - Treatments subjected to 8 days of 30°/15°C were then transferred to 35°/20°C for a further 7 days.

2 - All other abbreviations are listed on page 510

TABLE A3.19

CUMULATIVE GERMINATION SCORES OF SEEDS OF EACH COLLECTION IN EXPERIMENT 13, TRIAL 1

d 0											ty				
Collection	Sub-sample	Replicate		Day (Da	rà c	) =		l No	ven	nbei	of to:,	ria 1968 10	1 3) 11	12	Total viabilit
<u>ဗိ</u>	<u>အ</u>	<u> </u>	1_	2	3	4	5	6	7_	8					47
P2	1	1 2 3 4	0 0 0	0000	0 1 0 0	0100	0 1 0 0	0110	0110	0110	0110	0 1 1 0 0	0 1 1 0 0	01100	50 49 49
	2	1 2 3 4	0 0 0	0000	0 0 0	0 0 0	0010	0000	0020	0 0 2 1	0 0 2 1	0 0 2 1	0 2 1	0 2 1	50 49 49 49
	3	1 2 3 4	0 0 0	0000	0 0 0	0 0 0 0	0 0 1 0	0 0 0 1 0	0 0 1 0	0 0 0 0	0 0 1 0	0 0 1 0	0010	0 0 1 0	47 50 49 51
	4	1 2 3 4	0 0 0	0 0 0	0 0 0	0 0 0 0	0 0 1 1	0 0 1 1	0 0 1 1	0 0 1 1	0 0 1 1	0 0 1 1	0 0 1 1	0011	51 50 49 47 16
P3	1	1 2 3 4	0 0 0	0 0 0	0 0 0 0	0000	0 0 1 0	0 0 1 0	0020	0 1 2 0	0 1 2 0	0 1 2 0	0120	0 1 2 0	16 16 14
	2	1 2 3 4	0 0 0	0 0 0 1	0 0 0 1	0001	0 0 0 1	0 0 0 1	0 0 0 1	0 0 0 1	0 0 0 1	0 0 0 1	0 0 0 1	0 0 1 1	45 45 50 48 48
	3	1 2 3 4	0 0 0	0 0 0	0 0 0	0 0 0	0 1 0 0					0112	0 1 1 2		50 55 45 49 48 50
	4	1 2 3 4	0	0 0 0	0000	0 0 0	00001	0 0 0 1	000	0 0 0 1	0 0 1 1	0 0 1 1	0 0 1 1	0 1 1	45 48 50

Note: Abbreviations are listed on page 510

TABLE A3.20

9	otal tability		\$ 600°C	2722	0244 07480	0 4 4 4 0 0 0 0 0	2222	24 L 2	7000	7222
2 TO		4	0707	0400	0770	0000	00%	707	0000	L00
TRIALS		13	W-0-	0400	0770	0000	00WL	7077	0000	40 V
, TR.		12	707	0400	0770	0000	00%	7707	0000	707
T 13	trial	7	707	0470	0770	0000	00%	7707	0000	707
experiment	44	9	7707	0470	0000	0000	7000	7077	0000	707
XPER	o Su	6	707	0470	0700	0000	7000	7077	7000	707
IN I	beginning	80	rr0r	0470	0700	0000	7000	7077	0000	707
		7	<b>~~00</b>	0440	0700	0000	2000	7077	0000	707
TREATWENT	from	9	<b>~~00</b>	0470	0700	0000	00UT	7077	0000	707
	Days	5	<b>7000</b>	0470	0700	0000	0077	7707	0000	707
EACH	А	4	<b>6000</b>	0470	0700	0000	0077	7077	0000	700
S OF		2	7000	0470	0700	0000	0077	7077	0000	700
SEEDS		7	0000	0000	0000	0000	0007	0000	0000	001
10		~	0000	0000	0000	0000	0000	0000	0000	00
SCORES	plicate	Кеј	∠0w4	<b>し</b> るる4	70m4	<b>-</b> 6004	- LUN4	- CUW4	<b>ころり</b>	- 70
•	lo satisa Lorescend		원 +	<u></u> 다	<del>ዛ</del>	댐	+ R	띡	+ 교	<b>4</b>
MINATI	noitis Jusíq		09+		+ 7 7		00+		-15	
E GEF	moitoeL.	Col	P2							
CUMULATIVE GERMINATION	e trial begun (O <b>v</b> ab	wss	12-12-68							

- 4044 0000 4004 0000 mpath 0000 more 0010 012 - -0-- 0000 -001 00mm mata 000m annt 0000 0--- 00-0 at L LOLL 0000 LOLL OLWU L400 0L00 0U44 0000 0LLL 00L0 L2 ~ LOLL 0000 LOLO OLWU L400 OLOU RU44 0000 0000 LC L LOLL 0000 LOLO OLWU L400 OLOL RU44 0000 0000 CC L LOLL 000L LOLO 0LWU L400 0L0L @U44 0000 0000 C( L LOLL 0000 LOLO 0LWU L400 0LOL NU44 0000 0000 0000 0 14 4044 0000 4040 04WU 4400 0404 NU44 0000 0000 0000 0 q 꾸 떢 범 9

54 WUSE <u>7455 5514 8885</u> 2285 2287 566 5587 -- 00-0 05-WL 0-0W 0840 -0-0 4W0W 4-0- 4-64 LL 0010 ULWL 010W 5840 LV10 4W0W 4101 456W -LL 00L0 L4WL 000W D840 LDL0 4W0W WLOL W06W 00 0000 LUWL NOON 5040 LCLO 4WUN NLOO LCLW 000 0000 LUWL 10000 10040 LVL0 4WUW WL00 LV40 000 0000 LULO NOOU L® + 8 LVLO 4 NUW LOOO LNLU 000 0000 0770 NOON 0870 0570 4NUN 7000 7577 000 0000 0770 NOON 0870 0570 4Myw 6000 600 000 0000 01-0 NOON RBLY 05-0 4myw 0000 5NG-000 0000 0+00 r000 para 0000 4mgm 0000 c46c 000 0000 0000 n000 0000 0000 4mgm 0000 00nvo 000 0000 0000 4000 0000 0000 4 man 0000 0000 000 0000 0000 0000 0000 0000 0000 0000 000 0000 0000 0000 0000 0000 0000 0000 サタフレ かんろし かんろし かんろし かんろし かんろし かんろし かんろん 떢 ਸ਼੍ਰ 侔 년 4 벞 460 460 **P**3 P2

Totāl Visdility	2024 2024	\$57.5	. 2222	2024 2022	05448 8448	44 70 70 70 70 70	144W II	52.
4	0000	~0~0						
<u> </u>	0000	0000						
7	0000	0000	N <b>−</b> N N	22272	900	3~-8,		<b></b>
La 7	0000	0000	W <b>~</b> U O	2272	900	24 -62		~ ~
: trial	0000	0000	~~~~~	727	ω <sup>2</sup> 002	6,4 -8		~~
႕ o ရွာ	0000	0000	01-010l	4 1 2 2 2	NO 80	4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4		<b>6</b> 0
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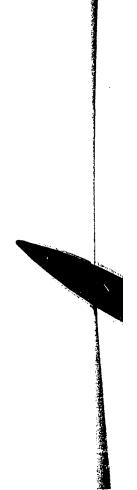
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#### APPENDIX 4

#### ANALYSIS OF VARIANCE

A4.1 Analysis of variance when the data are proportions General

The statistical technique known as Analysis of Variance (ANOVA) depends upon two assumptions: (1) that the observations  $\mathbf{x_{ij}}$  within any class j are normally distributed with mean  $\mathbf{p_j}$  and variance  $\mathbf{\sigma_j}^2$ ; and (2) that the variance  $\mathbf{\sigma_j}^2$  is constant from class to class.

Analysis of a two-way or factorial design in which these assumptions are not met cannot be handled by the conventional algorithm. When the data are of a non-normal distribution or when the error variances are heterogeneous, the ANOVA algorithm tends to produce significance by "F" or "t" tests for too many differences.

### Binomial data

Observations that are of the nature " $\underline{r}$  successes out of  $\underline{n}$  trials" are characteristic of a binomial distribution that is non-normal in nature and in which each proportion  $\underline{p}$  has a variance  $\underline{p(1-p)/n}$ , where  $\underline{p} = \underline{r/n}$ . When observations are replicated within one class, differences between replicates constitute another source of variance (Cochran, 1943). Thus the total variance of a proportion,  $\underline{V(p_i)} = \underline{p_i(1-p_i)/n_i} + \sigma_i^2$ , has two components where the first term represents

binomial variance and the second term extraneous variance.

Note that the binomial variance is dependent in part on the number of trials that constitute an observation and in part on the proportion of successes.

Heterogeneity of the variance that arises from differences in the component of binomial variance can be corrected in non-extreme cases by transforming the data to When heterogeneity is a function of a different scale. the component of extraneous variance remedial action is more complicated and involves weighting. Situations in which the amount of extraneous variance differs widely between classes are to be expected in analysing systems that by nature start from zero percent successes and ultimately reach one hundred percent. In germination trials for example, extraneous variance will be zero before germination commences and after the completion of germination in all replicates. During the period in which germination is taking place replicates will differ and a maximum value of extraneous variance will occur.

# Determining the nature of the variance

Cochran (1943) describes methods for determining the relative amounts of binomial and extraneous variance and for determining the efficiency of different types of weighting. These techniques apply only to data that conform to a one-way classification. No method is specifically provided for factorial designs. However, if the purpose of weighting is to assist in stabilising the variance, it is possible to

assess the value of weights and transformations by observing their effect upon the variances that estimate the error variance. Such an assessment can be made by performing Bartlett's Test of the homogeneity of variance both before and after transforming the data or applying weights.

#### Transformations

Snedecor and Cochran (1967 section 16.8) describe three transformations that may be performed upon binomial data.

- (1) The proportions can be regarded as normal variates and weighted by the inverse of their binomial variance, i.e.  $w_{ij} = n_{ij}/p_{ij}(1-p_{ij})$ .
- (2) The proportions can be transformed to equivalent angles; this is the arcsin transformation. The variance of a transformed value is approximately  $821/n_{ij}$  and thus as long as the  $n_{ij}$ 's are constant, further analysis does not require weights. The transformation is to the angle  $\propto$  (in radians) whose sine is  $\sqrt{p_{ij}}$ . When  $n_{ij}$ 's are less than 50, a modification of the transformation is advised when  $p_{ij} = 0$  or 100%. Mosteller and Youtz (1961) present a more complex transformation equation that takes into account the size of  $n_{ij}$ :

y<sub>ij</sub>=½(arcsin/(p/n+1) + arcsin/(p+1/n+1)).

The arcsin transformation may also be used with proportions that are subject to extraneous variance if this is of the form  $C_{ij}^2=k\cdot p_{ij}(1-p_{ij})$ , where k is a constant. This is often the case when the extraneous variance is

greatest with proportions around 50% and smaller towards the extremes of the range (Cochran, 1943, Snedecor and Cochran, 1967 section 11.16).

(3) The third transformation that is available is the logit transformation where:

$$Y_{i,j} = \log_e(p_{i,j}(1-p_{i,j}))$$

with variance:  $V(Y_{ij}) = 1/(n_{ij} \cdot p_{ij}(1-p_{ij}))$  and thus would require weights  $w_{ij} = n_{ij} \cdot p_{ij}(1-p_{ij})$ .

### Choice of transformations

When all  $p_{ij}$  lie between 25% and 75% the effects of these three transformations seldom differ. This is because the function  $p_{ij}(1-p_{ij})$  is approximately constant within this range.

When p<sub>ij</sub>'s extend over a wide range from near zero to 50% or higher there are reasons to experct that factorial effects will be more nearly additive on a logit scale than on the original scale of proportions. Logits extend the scale at both ends of the range (0 and 100%) so that the new range extends from -∞ to +∞. Thus with logits it is possible to avoid interactions that are purely a consequence of the original scale.

Arcsins are intermediate between logits and proportions in their effect. The scale is extended at both extremes, but remains finite within the range 0° to 90°.

A further factor influencing the choice of a transformation is the nature of the experimental design and the choice of analytical techniques that this design will permit. With

factorial designs, the analysis of weighted observations becomes complex.

With a one or two-way classification (where interactions are not considered and logits inappropriate) the decisions about weighting and transforming can be made independently following the criteria and procedures described by Cochran (1943). If the decision is then made to weight observations, the analysis of variance is easily computed; it is essentially similar to the analysis of unweighted observations for unequal numbers of observations per class.

With a factorial classification there are problems of orthogonality in performing a weighted analysis. These problems are the same as those encountered in a factorial analysis with unequal replication. In both of these cases, the interaction terms are strongly influenced by the distribution of replicates or weights (Snedecor and Cochran, 1967 chapter 16). With unequal replication, the means are weighted with the number of observations making up the mean and in the most unbalanced situations, a Least Square Analysis is performed. In such a case there is no provision for random effects. With individual observations weighted the appropriate model would need to include random effects. Cochran (pers. comm.) states that no such model has yet been devised.

With a randomised block structure blocks could be included as an additional factor and a least square analysis made with one weighted observation per cell in a factorial

design of one dimension greater. The effects would now include interactions between blocks and each other factor, terms that are normally absent but conceivably of importance. In a three-factor plus block design there would also be one four-factor interaction term, four three-factor interaction terms and six two-factor terms. The meaning of these terms would require much thought and the mechanics of the analysis would need to be derived from the three-factorial model which is sufficiently complex in itself.

None of these complications need be considered if the deviations among the subclass number  $n_{ij}$  are not excessive, or if they are distributed proportionally among the factors. Also, if the amount of variance contributed by unequal numbers is small relative to that from extraneous sources, weights based on sub-class numbers are of no value at all. When sub-class numbers are equal within the levels of one factor or proportional within the levels of two or more factors, the methods described by Snedecor and Cochran (1967 sections 16.3 and 16.4) for unequal replication are easily modified.

## Summary of analysis procedures

## 1) One way classification

Decide whether an arcsin transformation is appropriate. If sub-class numbers are equal an unweighted analysis is performed. If sub-class numbers vary widely a test can be made of the efficiency of equal weights within either

treatments or replicates and of the efficiency of an unweighted analysis. The effect of the weights should again be examined by Bartlett's test.

## 3) Factorial classification

Where treatment replications are not included the choice of transformations can be made without concern that a weighted analysis may be necessary. Least square analysis is suited to these situations (Snedecor and Cochran, 1967, Chapter 16) or analysis may be performed using information theory (Kullback, 1959).

Where treatments are replicated, the choice of transformation excludes logits since these require weights that are not likely to be proportional within the different If sub-class numbers are equal any of the methods factors. of unweighted analysis of variance described by Snedecor and Cochran (1967 chapter 12) may be used. If the sub-class numbers vary widely but are proportional within factors the weighted analyses described by Snedecor and Cochran (1967 The use of Bartlett's test both chapter 16) may be used. with and without weights indicates whether the weights used Techniques have any stabilising effect upon the variance. are now available (Orloci, pers. comm.) whereby information analysis can be used with replicated observations within treatments.

Figure A4.1 outlines the sequence of analysis that is exployed with proportional data in a factorial classification.

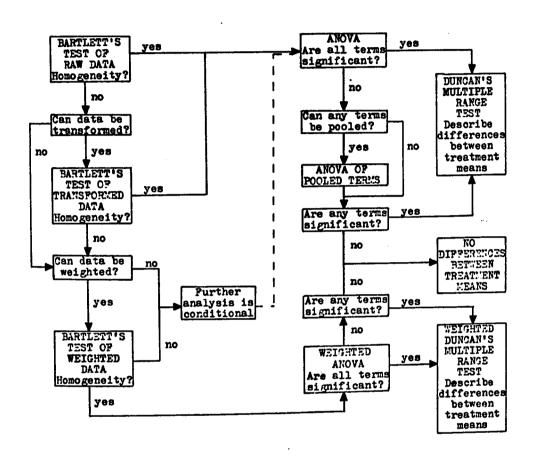


Fig. A4.1 Stages in the analysis of factorial designs of data that are proportions

## Conditional analysis

In the event that weights and transformations do not achieve stabilisation of the variance, it is possible to continue with the analysis of variance, bearing in mind that one of the basic assumptions does not hold. As mentioned previously, the effect of heterogeneity of variance is to cause too many differences to be declared significant by F-tests or t-tests. However, Snedecor and Cochran (1967 section 11.13) state that this effect is only mild when the replication of treatments is equal.

A4.2 The analysis of nested or hierarchical designs

A nested nalysis is required when the treatment (or fixed effect) that is being compared is represented by samples which are themselves represented by sub-samples. For example, to compare the germination of two or more species it may be decided to sample several geographically isolated populations to represent each species, to subsample several plants of each population to represent the population and to sub-sub-sample several seeds (or batches or seeds) to represent each plant. Plant one of population one bears no logical relationship to plant one of population two and thus it would be incorrect to analyse these results in a factorial design.

#### Unbalanced designs

When all the samples are of equal size, all the subsamples are of equal size and all the sub-sub-samples are of equal size, the nested analysis of variance is easily performed (Snedecor and Cochran, 1968 section 10.16). If the sample (sub-sample etc.) numbers are unequal the analysis of variance becomes more tedious. Snedecor and Cochran (1968) describe a technique that evaluates unbiased estimates of the variance of each level of the hierarchy. The expected value of the variance of each level contains a component of variance of that level and also a component of the variance of each subordinate level. In the example described above, the expected population variance includes a component of population variance and components of plant In the balanced design (i.e. with equal and seed variance. sample sizes) the coefficient of the variance of a particular level remains the same in all levels in which that component This is not the case with an unbalanced is included. design and consequently it is no longer possible to use a ratio of variance estimates to test the null hypothesis of zero variance at any level.

#### Approximate 'F' test

Tietjen and Moore (1968) have outlined both the theory and mechanics of an approximate 'F' test that can be used to test the null hypothesis of zero variance at each level. In order to compare this approximate variance ratio with tabulated critical values it is also necessary to calculate a value for the degrees of freedom of the denominator for each level.

## Comparison between the unnested means

All but the highest level of the hierarchy represent

random sampling and although it is informative to test for variance within each level there is no logical reason for comparing the means of samples (and sub-samples etc.). Moreover there is probably no model that would permit such comparisons were they required (Tietjen, pers. comm.) When the highest level in the hierarchy is a fixed effect then there are valid, logical reasons for comparing the For example, in a comparison of several species one means. might wish to know which species were significantly different Eisen (1966) describes a procedure for in their response. determining the variance and hence the standard error of each mean of the highest (unnested) fixed effect. this parameter it is then possible to perform a suitable test of the means, e.g. Duncan's new multiple range test (Steel and Torrie, 1960 section 7.5).

#### APPENDIX 5

RESULTS OF STATISTICAL ANALYSES OF GERMINATION DATA A5.1.Introduction

The following tables present the results of the statistical analyses of experiments 9 to 13. Each stage in the analysis of one experiment (or germination trial) is described before the next experiment is presented, and within each experiment a similar sequence of stages occur. These are described under the following headings:

## 1) Statistical design

For most of the analyses the data was handled by an IBM 7040 computer using programmes written in the ALGOL language (modified for the University of Western Ontario, Department of Computer Science ALGOL compilers). In using these programmes each experiment was described in terms of Factors A, B and C and this standardisation provided a useful abbreviation for presenting the results. Details of the design code used to describe each experimental factor are presented for each experiment under the above heading. The information is presented in two ways depending upon whether the design is factorial or hierarchical. The descriptions for factorial designs are self explanatory. One anomaly that should be explained occurs with one or two factor designs. In a two-factor design the factors are

described as B and C rather than A and B as one might expect. In a one-factor design that factor is described as C.

The description of hierarchical designs is less obvious and an example is provided for explanation.

Consider experiment 9, germination trial 1, hierarchical design. Table A5.1 describes the following hierarchy:

design.	010 1-70			
Level Code:	0	1	2	3
Experimental Level:	Replicates	Sowings	Collections	Species
Categories:	1,2,3	<b>S1</b>	_	
	1,2,3	S3	P1	-
	1,2,3	<b>S</b> 1	_	
	1,2,3	S3	P2	_
	1,2,3	S1		
	1,2,3	<b>S</b> 2		
	1,2,3	S3		
	1,2,3	S4	P3	A. powellii
	1,2,3	S1		
	1,2,3	\$2		
	1,2,3	S3	R1	A. retroflexus
	1,2,3	s1	Н7	A. hybridus
Number of categories in each level:	36	12	5	3

In table A5.1 the column headed "Contribution of categories to the next level" can be illustrated with the above example for levels one and two. Beginning at the

top of the column for level one (above), the number of categories of level one that contribute to the first category of level two is two. The next two categories of level one contribute to category two of level two and the next four contribute to the third category and so on. At the same time each category of level one consists of three categories from level zero.

# 2) Bartlett's test and the analysis of variance

The results of these two analyses are presented in either separate tables or a combined table depending upon whether the effects of weights and transformations have been examined. If several Bartlett's tests have been performed on one set of data to determine the most appropriate analysis then the results are recorded in a separate table. Otherwise the results of Bartlett's test and the analysis of variance are combined.

In presenting the results of the analysis of variance only the variance ratios for each effect are reported. In factorial analyses in which the degrees of freedom remain the same for day to day analyses of the same design, it has been possible to include critical values (from the tables in Snedecor and Cochran, 1968) upon which the tests of significance are based. In the unbalanced nested design the denominator degrees of freedom for each variance ratio vary independently of the design and in these cases only the degrees of freedom and a comment on significance have been included.

# 3) Duncan's multiple range test (see Harter, 1960)

In a factorial experiment range tests are conducted for each factor in the design. Within a factor that is to be tested, the range tests are repeated for each combination of levels of every other factor. The means of each level of the factor under consideration are assembled in rank (from highest to lowest) and the highest mean is compared with each of the subordinate means. Then the second highest mean is compared with its subordinate means and so on. C is the number of levels of the factor being tested then within any one combination of the levels of the other factors there will be C(C-1)/2 comparisons. For a three factorial design with five levels of factor one, three levels of factor two and two levels of factor three there are 105 individual comparisons to be made for each day of the germination trial. For this reason the critical and observed values compared in each range test have been omitted and the results are presented in summary form only. table is included for comparisons within each factor that shows a significant variance ratio for at least one day during the germination trial. In each comparisons within the table the days are listed for which the two means being compared are significantly different and the direction of this difference is also noted. This is in the form (for example):

P1 P2:1-6 (The symbol & represents "greater than")

In this example the germination total for collection P1

significantly exceeds that for collection P2 from days 1 to 6.

The tables take two forms depending upon the number of levels in the factor they describe. There the factor is represented by two levels the table is broken into blocks that represent the levels of the other two factors (in a three factorial design) in a two dimensional matrix. There the factor contains more than two levels the table consists of a half matrix of blocks of each of the C(C-1)/2 comparisons within that factor. Cells within each block represent the levels of the other two factors in two-dimensional sub-matrices.

# Abbrevi tions used in Appendix 5

Weights

EW

equal weights = unweighted

WA, WB, WC

weights proportional within one

factor; A, B or C

WAB, WBC, WAC

Weights proportional within two

or AB, BC, AC factors

WABC or ABC

Weights proportional within three

factors

Transformations

TRANS

Transformation

17

Freeman-Tukey arcsin transformation see Mosteller and Youtz (1961)

Bartlett's test and the Analysis of Variance

DF or d.f

Degrees of freedom

DFN

Mumerator degrees of freedom

DFD

Denominator degrees of freedom

TX

Degrees of freedom exceed 1000

Chi<sup>2</sup>

Chi-squere (X2)

А, В, С

Main effects A, B and C

AB, EC, AC,	Two-factor interactions AxB, BxC, and AxC
ABC	Three factor interaction AxBxC
SIG	Significance of observed value
* *	Significant at the 1% level of probability or lower
*	Significant between the 5% level of probability or lower
ns	not significant at the 5% level of probability or lower
p=0.05	5% level of probability
p=0.01	1,3 level of probability
VR	Variance ratios
REPS	Replicates

# Duncan's Hultiple range tests

"greater than" (equivalent to the conventional symbol >)

Other abbreviations used for factor levels in these tables are the same as those used in Appendix 3, on page 510.

### General abbreviations

TO AT 21 021 0210	12 toto1
SS	Proportions based on the total number of seeds sown per replicate
TG	Proportions based on the total germination (ultimate) per replicate

A5.2 The analysis of data from experiment 9

# A5.2.1 Germination trial 1

### TABLE A5.1

# THE ANALYTICAL DESIGN OF THE HIERARCHICAL ANALYSIS OF DATA FROM EXPERIMENT 9, TRIAL 1

Level	Experimental level	Number of categories	Contribution of categories to next level
0	Replicates	36	3 to each category of level 1
1	Sowings	12	(2,2,4,3,1,) to level 2 categories respectively
2	Collections	5	(3,1,1) to level 1 categories respectively
3	Species	3: A. pow A. hyb	ellii, A. retroflexus and ridus.

# TABLE A5.2

# THE RESULTS OF BARTLETT'S TEST AND THE HIERARCHICAL ANALYSIS OF VARIANCE PERFORMED WITH THE DATA FROM EXPERIMENT 9, TRIAL 1

Chi <sup>2</sup> SIG	Leve sowi	 STG	coll VR	evel ecti DFN	ions	SIG	:	Leve spec DFN	ies	SIG
							1.10	2	3	ns

Mote: Abbreviations are listed on page 556.

TABLE A5.3

STATISTICAL DESIGN OF THE FACTORIAL ANALYSIS
OF DATA FROM EXPERIMENT 9, TRIAL 1

Factor code	Experimental factor	No. of levels	Levels
В	Sowings	2	Sowing 1 and Sowing 3
C	Collections	3	P1, P2, P3
Mote:	Abbreviation	s are 1	isted on page 556.

TABLE A5.4

THE RESULTS OF BARTLETT'S TEST AND THE FACTORIAL ANALYSIS OF VARIANCE OF DATA FROM EXPERIMENT 9, TRIAL 1

Item	В	C	BC	REPS	ERROR
DF	1	2	2	2	10
Critical values: p=0.05 p=0.01	4.96 10.0	4.10 7.56	4.10 7.56	4.10 7.56	
Observed values	0.02	16.0	Q.60	0.36	
SIG	ns	**	ns	ns	

Note: Abbreviations are listed on page 556.

# Duncan's multiple range test of the means

As neither sowing time nor the interation of sowing time and collection gave significant variance ratios, the values for the two sowings were pooled to give one mean for each collection. These means were compared using Duncan's new multiple range test. Table A5.5 presents the results of this comparison.

### TABLE A5.5

THE RELATIONSHIPS BETWEEN COLLECTION MEANS AS DETERMINED BY DUNCAN'S MULTIPLE RANGE TEST OF DATA FROM EXPERIMENT 9, TRIAL 1

	P1	P2
P2	P1=P2	
P3	P3 <b>≬</b> P1	P3≬P2

Motes: 1 - ≬ = "greater than"

2 - Other abbreviations are listed on page 556.

# TABLE A5.6

STATISTICAL DESIGN OF THE ANALYSES OF DATA FORMING EACH COMPARISON IN EXPERIMENT 9, TRIAL 3.

Factor code	Experimental factor	No. of levels	Levels
A B	Collections Wintering sites	4 3	F1, P2, P3, R1 Buried, Surface, Laboratory Harvest 1, Harvest 2
С В С	Collections Wintering	5 4	P1, P2, P3, R1, H7 Buried, Surface, Laboratory, On-plant
<b>А</b> В С	Collections Sowings Wintering sites	2 2 2 4	P2, P3 Sowing 1, Sowing 3 Buried, Surface, Laboratory, On-plant
A B C	Collections Sowings Wintering sites	3 2 3	P2, P3, R1 Sowing 1, Sowing 3 Buried, Surface, On-plant
B C	Collections Wintering sites	3 3	P2, P3, R1 Buried, Surface, On-plant
	C B C A B C B C	A Collections B Wintering sites C Harvests  B Collections C Wintering sites  A Collections B Sowings C Wintering sites  A Collections B Sowings C Wintering sites  B Collections B Sowings C Wintering sites  C Wintering sites	A Collections 4 B Wintering 3 sites C Harvests 2 B Collections 5 C Wintering 4 sites  A Collections 2 B Sowings 2 C Wintering 4 sites  A Collections 3 B Sowings 2 C Wintering 3 sites  B Collections 3 C Wintering 3 Sites

(continued)

Compa- rison	Factor code	Experimental factor	No. of levels	Levels
9/5	A B C	Collections Harvests Wintering sites	3 2 2	P2, P3, R1 Harvest 2, Harvest 3 Buried, Surface
9/6	A B C	Collections Sowings Wintering sites	2 2 3	P3, R1 Sowing 2 Buried, Surface, Laboratory
9/7	В С	Sowings Wintering sites	2 2	Sowing 3, Sowing 4 Buried, Surface

Mote: Abbreviations are listed on page 556.

# 15.2.2.1 Analysis of viabilities

### TABLE A5.7

WEIGHTS USED IN ATTEMPTS TO STABILISE THE VARIANCE OF CERTAIN COMPARISONS AMONG THE VIABILITY DATA OF EXPERIMENT 9, TRIAL 2

Factor within Weights for each level of the which weights factor within which they are Comparison are applied applied Buried, Surface: 50, 9/1 Laboratory: 100 Buried, Surface, On-plant: 50 9/2 C Laboratory: 100 Buried, Surface, On-plant: 50 9/3A C Laboratory: 100

Mote: 1 - The weights were determined from the total numbers of seeds used in each treatment.

2 - Abbreviations are listed on page 556.

TABLE A5.8 RESULTS OF BARTLETT'S TESTS OF THE VIABILITY DATA FORMING EACH COMPARISON IN EXPERIMENT 9, TREAL 2

Compa-	DF	Criti values chi p=0.05	for E	Unweigh observed value			thted observed value	SIG
9/1 9/2 9/3A 9/3B 9/4 9/6 9/7	23 19 17 17 51 3	35.2 70.1 25.0 27.6 11.1 19.7 7.81	41.6 36.2 30.6 33.4 15.1 24.7	92.1 68.6 31.6 29.7 9.1 4	**  **  ns  ns	WB WC WC	91.9 62.8 33.6	** **
Mote:	۸h	hrevist	ions ar	e listed	on p	<sub>ag</sub> 556.		

Abbreviations are listed on page 10.

In comparisons 9/1 and 9/2 weighting the data reduced the heterogeneity of the variance slightly without achieving stabilisation. In comparison 9/3, weighting the data served to increase the heterogeneity of the variance. Thus a weighted analysis of variance was used for comparisons 9/1 and 9/2 and an unweighted analysis in all other comparisons.

TABLE A5.9

RESULTS OF THE ANALYSIS OF VARIANCE OF EACH COMPARISON AMONG THE VIABILITY DATA OF EXPERIMENT 9, TRIAL 1

Comparison	Weights	Item	A	В	C	АВ	BC	AC	ABC	REPS	ERROR
9/1	WB	DF VR SIG	3 1.09 ns	2 0.12 ns	1 0.23 ns	6 1.96 ns	2 1.81 ns	3 1.54 ns	6 0.55 ns	2 1.48 hs	46
9/2	MC	DF VR SIG		4 2.49 ns	3 16.5 **		12 6.08 **			2 0.57 ns	38
9/3A	EW	DF VR SIG	1 85.8 **	1 8.81 **	3 40.7 **	1 7•33	3 3.01	3 25.5 **	3 0.17 ns	2 0.15 ns	30
9/3B	EW	DF VR SIG	2 72.0 **	1 2.22 ns	2 51.7 **	2 3•45	2 1.07 ns	4 16.6 **	4 8.88 **	2 0.91 ns	3 <del>4</del>
9/4	27	DF VR SIG			2 13.3 **		2 1.57 ns			2 2.90 ns	10
9/6	EW	DF VR SIG	1 0.31 ns	1 0.32 ns	2 1.66 ns	1 2.49 ns	2 0.61 ns	2 1.14 ns	2 1.56 ns	2 0.63 ns	22
9/7	Ε'n	VR SIG	-	1 62.3 **	ns		1 1.83 ns			2 3.91 ns	6
$\mathbb{N}ot\epsilon$	<u>:</u>	Abbre	viatio	ns ar	e lis	sted c	n pag	$_{\rm se}556$	•		

# Duncan's Multiple range test of the means

As mentioned on page 258, the analysis of differences in viabilities is only of interest in the comparisons between wintering treatments. Thus table 5.10 (which includes several sub-tables) presents results for these comparisons

only. Each block within a sub-table is divided into 12 cells, arranged in three columns and four rows. The columns represent harvest times (harvest 1 to 3, from left to right) and the rows represent sowing times (sowings 1 to 3, from top to bottom). Each collection is represented by a separate sub-table.

TABLE A5.10

THE RELATIONSHIP BETWEEN MEAN VIABILITY OF SEEDS SUBJECTED TO DIFFERENT POST-HARVEST CONDITIONS

### a) Collection Pl

a) OOTIECUI	Buried	Surface	On-plant
	B=S B=S		
Surface			
	B≬P	S=P	
_			
On-plant			
·	<del>========</del> B=L B=L	S=L S=L	P=L
Laboratory			

# b) Collection P2

s/ <u></u>										
	•	Burie	đ	St	rface	Э	On-plant			
	B=S	B=S								
Surface		B=S	B=S							
	====	B=P		85,52	S=P	<u></u>				
On-plant		B=P	B=P		S=P	S=P				
٠	==== B=L	B=L		S=L	S=L		P=L			
		<del> </del>								
Laboratory		B=L			S=L		P=L			
c) Collection P3										
		 Buri	eđ	Ω	Surfac	ce	On-plant			

O) DOTTOODE	<u> </u>	4					
	]	Burie	d	Sı	ırface	9	On-plant
	B=S	B=S					
Surface	B=S	· · · · · · · · · · · · · · · · ·					
		B=S	B=S				
			B=S				
		B≬P			S≬P		
		·					
On-plant		B≬P	B=P		S≬P	s <u>.</u> P	
	B=L	B=L		S=L	S=L		L≬P
Laboratory				-			
				<del> </del>			
			=====	<u> </u>			

# d) Collection Rl

	<u>.</u>	Burie	đ	Sı	ırfac	е	On-plant
·	B=S	B=S					•
	B=S						
Surface		B=S	B=S				
		B=P			S=P		
On-plant		в≬Р	B≬P		S≬P	S≬P	
	B=L	B=L	<u> </u>	S=L	S=L		P=L
Laboratory							

### e) Collection H7

Sowing one, Harvest two: No significant differences in viability between seeds after different post-harvest treatments. These are the only values for this collection.

Note: Abbreviations are listed on page 556.

# A5.2.2.2 Analysis of daily germination totals

Table A5.11 indicates the weights that were used in attempts to stabilise the variance of each comparison. The weights were derived from the numbers of viable seeds per treatment averaged within each level of a particular factor. Subsequently Bartlett's test was performed for each comparison both with and without weights. In those comparisons in which weighting gave a lower chi-square a weighted analysis of variance was used.

TABLE A5.11

### WEIGHTS USED IN ATTEMPTS TO STABILISE THE VARIANCE OF COMPARISONS AMONG THE GERMINATION DATE OF EXPERIMENT 9 TRIAL 2

Comparison	Factor(s) within which weights are proportional	Weights for each level of the factor chosen
9/1	В	Buried, Surface: 49, Laboratory: 97
9/2	C	Buried, Surface: 49, On-plant: 41, Laboratory: 98
9/3A	C	Buried: 48, Surface: 47, On-plant: 35, Laboratory: 96
9/3B	A	P2: 49, P3: 38, R1: 47
	or C	Buried: 48, Surface: 47, On-plant: 38
9/4	В	P2: 49, P3: 26, R1: 46
9/5	В	P2: 50, P3: 36, R1: 49
9/6	C	Buried: 48, Surface: 47, Laboratory: 97

Note: Abbreviations are listed on page 556.

### Homogeneity of variance

The variance of raw data was examined using Bartlett's test in comparison 9/1 only. In this comparison transforming the data to the arcsin scale reduced the homogeneity chi-square for each day of the germination trial. This scale was thus to be preferred to the original scale of the raw data. In all of the other comparisons the arcsin transformation was applied to the data without a prior examination of the variance of the raw data. The results of Bartlett's tests are presented in table A5.12.

TABLE A5.12

THE RESULTS OF BARTLETT'S TESTS OF THE COMPARISONS AMONG THE GERMINATION DATA OF EXPERIMENT 9, TRIAL 2

SIG	* * * * * * * * * * * * * * * * * * *	* * * * * * * * * * * * * * * * * * *
ted data chi	00000000000000000000000000000000000000	77844440000040808 0000000000000000000000
Weights of		
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ormed chi2	00000000000000000000000000000000000000	97844450000000000000000000000000000000000
Transformed TRANS chi2	ほ复取鼠取取瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦	公瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦
date SIG	* * * * * * * * * * * * * * * * * * * *	
Raw chi2	20111110001100001 1011110001100001 1011110001100001	
tical values p=0.01	44.00	36.2
Critical chi2 values p=0.05 p=C	23.52	ν. •
DF	23	φ -
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Compa- rison	1/6	9/2

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75 71 86	404 <i>WWWW</i> 444 <i>VVV</i> 0011 188418 <i>V</i> 00444 <i>V</i> 4 <i>V</i> 0000 <i>W</i> 0	4 C & 4 WWWWW4 R R & W & C W & 4 R R R R R R R R R R R R R R R R R R
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	27.6	25.0
	17	7
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	9/3A	9/3B

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Weighted Weights	WA WA WA WA	WB WB WB WB WB WB WB		
data SIG	* * * *	* * * * * dddddddddd* * * * * *	* * * * * * * * * * * * * * * * * * *	
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Transformed TRANS chi <sup>2</sup>	<b>표</b> 표 표 대	<b>承环京京京京京京京京</b> 京京	瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦瓦	
cal ralues p=0.01		0 7	24.7	24.7
Critical chi <sup>2</sup> values p=0.05 p=0.0		75.5	19.7	70 0
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Compa-		9/4	9/5	U/ U

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Ö	) C	Q A	WC	MC	DI.	MC	NC.	WC	WC	WG	WC	MC	D M	NG.	WC	WC	 
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9/6																	Note:

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# Analysis of variance

In performing the analysis of variance weights were used in each situation where Table 5.13 presents the weighting reduced the chi-square value in Bartlett's test. results of the analysis of variance for each comparison.

TABLE A5.13

THE RESULTS OF ANALYSIS OF VARIANCE OF COMPARISONS AMONG THE GERMINATION DATA OF EXPERIMENT 9, TRIAL 2

ERROR	4.6																				
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ABG	9	2.30	3.22	2.0			0 * *	k/* •*			\ <u>~</u> 70	• 70	• rc	) <b>a</b> [ !	7 T S S	• (/)	• (I)	• 01	• •	• Ø	1.9 ns
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BG	~	3.20	5.10	7.	ns 17	•	39°5	34.9	0 0 0 *	25.7	17.8	13.0	© •* ∪*	2.7	2 t 3 c 5 c	7 t	• (0)	7 0.2 ns	• Ω	• Q	6 0.1 ns
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s Item	VR SIG VR VR SIG SIG	DF Fat P=0.05 VE	VR SIG VR SIG VR SIG VR SIG VR SIG VR SIG VR SIG VR SIG
Teichts	EM EM EM	E E	EM EM EM
Dey	15 17 24	7	0 m 4 0 r 80 0 P 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7 7
Comparison		75/6	

	46
ps 0.5 0.5 0.0 0.0 0.0	0
ns 1.0 1.0 1.3 1.7 1.7	4 0 W W* O HW* 4* W* W* W* W* 0 HO
* W * W * 4 * 4 * * O * W * 4 * 4 * C O C 4	4 0 0 0 * 0 * 0 * 0 * 0 * 0 * 0 * 0 * 0
ns 14.2 14.0 14.0 14.6 15.6	0
ps + 05 + 05 + 05 + 05 + 05 + 05 + 05 + 0	0
* † * † * † * † * * † * * † * † * † * †	0
ns 0.2 00.4 ns ns 0.1	7 4 7 4* C*
* C * C * C * C * C * C * C * C * C * C	0
SIG VR SIG VR SIG VR SIG	16   PERDEBERERERERERERERE
EN EN EN	WA W
75 17 21 24	- 0 w 4 0 c 8 6 6 6 7 7 7
	9/3B

		į		
TRROR		C   C   C		
REPS	0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	2 2 3 63 23 23 23	2	2 2 7
VBC	0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	 		
υγ	0* C* C* C* (N* N* N* O* C* C* O* O* N* N* N* C*	i 1 1 1 1 1		
BC	0 4 0 4 4 7 4 7 4 7 4 7 4 7 4 7 4 7 4 7	3.01	- 4- 4- 4- 4- 4- 4- 4- 4- 4- 4- 4- 4- 4-	C C
A.B	<ul><li>ω* Ω α ο α ο α ο α</li><li>ω* ω* ο α ο α ο α ο α</li><li>ω* ω* ο α<td>   </td><td></td><td></td></li></ul>	 		
Ö	0 * 0 * U * U * U * 4 * C * 0 * C * U * W * O * 4 0 N 0 0 W	6 23	M d C d w d ω * C * C * ω * C * w * 4 * π * ω * ω * ω * ω * ω * ω * ω * ω * ω	
£λ	0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	2.63	<ul> <li>Λ* Ο 4 L* W 4 L 4 O 4 4* L* Λ* Λ* Λ* Λ*</li> <li>4* • κ0 O* • κ0 • κ0 • κ0 * Λ* Λ* Δ*</li> <li>* Γ Ο • Λ Γ 4 Λ • * Λ* Γ* 4* Γ*</li> <li>* Γ Γ Γ Γ Γ Γ Γ Γ Γ Γ Γ Γ Γ Γ Γ Γ Γ Γ Γ</li></ul>	
4	C* O* O* D* O* C* C* O* O* D* O* C* O 4 C O C O			
c Item	VR SIG VR VR VR SIG SIG	DH BBC BC 0.05 D=0.01	SIG VE SI	) 
Weight	WA WA WA WA		EW E	TP/II
Day	47 77 77 77 77 78		- u n 4 0 c 0 0 6 7 7 7 7	ر د
Comparison		₩/6		

						22													
ns 0.5	• U		) • U	າ • ທ	ປັ• ຕັ	2	3.44	5.72	1.7	ns o	7 • C	2.5	ns 1.6	น () • ภ	ns, 2.9	ns 2.0	ns 1.1	0 n s n s n s	
						2	3.44	5.72	0.7	n c	- 84 - 84	3.0	ns 7.1	ក្នុង ស្វ	ns. 0.	ns 1.4	ns 1•3	0 ps 0 s d 8 s	
						2	3.44	5.72	•	ທ .	• ທ	•	<b>ω</b> •	ns 0.2	Ω •	ns 0-	<b>Ω</b> •	n 0 n s 1 s 2 s 3 s 3 s 3 s 3 s 3 s 3 s 3 s 3 s 3	
* C.* 						     	4.30	7.94	4.0	ns OO4	) N	•	ns 0.5	<b>ω</b> •	ns 4.3	•	•	7.0 10.0	
					·	2	3.44	5.72	10.5	* 5	) ທ	•	ns 0.8	<b>ω</b> •	Ω •	ns 0.8	Ω •	ns 10.1 11.2	
* 0,* * 0,*		49.4	78.7	104.3	101.7	_	4.30	7.94	0.2	ns 7.9	*	1.0	ns • 04	0 n n n n n	ns 0.2	ns 0.7	ns 2.0	a 4. •	
** ***	20 10 10 10 10 10 10 10 10 10 10 10 10 10	0 8 8 *	41.0	ω * × * ×	** **	<u></u>	4.30	7.94	77.	+ O	O (	•	$\omega$	Ω •	Ω •	ທ •	ns 1.5	ns 0.5 ns	
						2	3.44	5.72	25.5	• •	· m	73.7		· * *	•	Ω •	U) •	n × √* • * • 8	
SIG VR SIG	VR SIG	VR	VR SIG	VR SIG	VR	۱ (	10 = 0 = 0 = 0 = 0 = 0 = 0 = 0 = 0 = 0 =	S C .	VR	or Car	SIG	VR 310	SIG VR	SIG Ve	SIG	SIG VR	VIR VIR OTS	VE VE SIG	
WB	H		WB	្រ	WB					MET.		125	M	A STATE OF THE STA		ME	17:E	157	
14	7	16	17	2	24				<b>~</b>	C.		M	4	Ø	2	ω	σ,	70	
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(continued)

ERROR		55
REPS	0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	2
VEC	0.04 0.03 0.03 0.03 0.03 0.03 0.03 0.03	2
VQ.	00000000000000000000000000000000000000	0
BG	D D D D D D D D D D D D D D D D D D D	2
ΛB	0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	4.30 4.30 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00
D	4* 4* 0* 00 * 10* 10* 10* 10* 10* 10* 10* 10	0 W R C * 4 * 8 * 8 * 8 * 8 * 8 * 7 * 8 * 8 * 8 * 8
Ħ	.04 .05 .05 .05 .05 .05 .001 .001 .001 .001 .001 .001	4.0 4.0 4.0 4.0 4.0 4.0 4.0 4.0
V	8 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	- 4 C 4 t 4 + 0 t 0 * C * 0 * C * 0 * 0 * 0 * 0 * 0 * 0 *
Item	VR SIG VR SIG VR SIG VR SIG VR SIG VR SIG	DF F at P at P at P at VR VR SIG VR SIG VR SIG VR VR SIG VR VR SIG VR VR VR VR VR VR VR VR VR VR VR VR VR
Weights		WG W
Day	27 77 77 75 42 42	700700000000000000000000000000000000000
Compertison		9/6

su	٠ 0	ns	ν., Ο	*	ง เก	អន	7.	ns	<u>_</u>	ដន	7.0	ns	0.7	ВS	ر ا ا	ns	7.0	มร	7.	ns	2.7	มร	<u>.</u> ر	ងន	0.	ns	0.3	ns.	0	มะ
us	٥ •	ns	2.4	ns	oj R	ns	9.0	ns	٥ 0	ង	7	ns	ω. Ο	ns	o. 0	ns	٠ <b>،</b>	ns	٥ •	ns	٠ •	ns	٥ •	ns	0.5	រាន	0.7	มธ	ง 0	ns
* *	7.	ns	0.3	ន្ត	<u>1</u> س	ns	7.4	ងន	о О	ns	0 0 0	ងន	0.7	ns	0.2	ns	0.3	ns	0.2	ns	0.4	ns	2.3	ដូន	3.5	DS	7.	ns	2.0	ns
su	0.5	ns	٠. ر	ns	٥.	ns	0.7	ns	٠. 5	su	9.0	ns	٠. ا	ងន	0.1	ns	0.7	ns	0.0	มร	9.0	ns	8. 7.	* *	10.7	* *	0.2	ns	0.7	ns
																													0.7	
*	39.8	*	56.7	*	72.1	*	86.6	* *	105.3	*	152.3	*	56.3	*	0.43	* *	22.0	*	16.3	*	15.0	\ <b>*</b>	8.3	*	7.9	* •*	S 0.	ns	3.3	ns
บร	ω 0	มร	0.3	ns,	0	ns	0.3	ns	۲. ۲.	มร	2	۲. ب	7	E	0.5	TIS.	0.4	มร	0.4	ns	0	ns	3.8	ns	7.0	*	0.2	ns	0.3	ns
\ ' *	20.00	ns	0	*	15.9	\ \ <del>*</del>	23.9	*	17.5	-* *	യ പ	\ • <del>*</del> • *	40.	თ დ	<u>-</u> ر	, S.C.	N N	118	1.7	ns.	4.0	SE	7	, w	7.1	- *	2.2	ងន	2.0	ns
SIG	VE	STC.	VE VE	む上の	VE VE	<b>む</b> に な	VR	STS	VR	<u>ئ</u> الله	) \ } }	<u>ئ</u> لــــــــــــــــــــــــــــــــــــ	) 1 (2)	せ と と い	7. A. V.	せ上の	VEN VEN	the second	<b>3</b> 1 (2: 2 (2:	STS.	VB C	1 T	VE V	STC	) R	ر الم	N EN	PIC DIC	VR.	SIG
)	MC	<b>)</b>		<b>)</b>	OI.	)	DL.	) :	MC	) :	ָרָי בּיִּ	) :	E	į		į	TAKH.	;	ME		HILL	į	PE-	į	Fig.	į			E	
7	70	`	77	•	Q	)	5	•	00	)	O	i\	7	2	7	J	ر لا	)	74	<u> -</u>	ر ر	`	77	2	70	<b>.</b>	7	- J	24	

Note: Abbreviations are listed on page 56.

### Duncan's multiple range tests

Multiple range tests were used to determine the significance of differences between the means in each comparison. The results of these tests are presented in table A5.15. Some of the comparisons included identical components. Rather than presenting the range tests for factors more than onec, the comparison that included the most levels of the factor was the only one included. Table A5.14 lists the comparisons that are duplicated.

TABLE A5.14

A LIST OF DUPLICATE COMPARISONS BETWEEN FACTOR MEANS AMONG THE GERMINATION DATA OF EXPERIMENT 9, TRIAL 2

Harvests	One	Sowings Two	Three
One	9/1 ( <u>BSL</u> <u>P1/2/3 R1</u>	9/6 ( <u>BS</u> R1	
	9/6 (BS 173 R1		
Two	9/1 (BSL (P1/2/3 R1		9/5 (BS 9/5 (P2/3 R1
	9/2 (BSLP P1/2/3 R1 H7		9/3 ( <u>BSLP</u> <u>F2/3</u> R1
	9/3 (BSLP 9/3 (P2/3 R1		
Three			9/4 ( <u>BSP</u> <u>P2/3 R1</u>
			9/5 (BS P2/3 R1
Notes:	1 - The comparisons chin table 5.15 are	e miner raneas	
	2 - Abbreviations are	e listed on pa	<sub>uge</sub> ბან.

### TABLE A5.15

THE RELATIONSHIPS BETWEEN MEAN PERCENTAGE GERMINATION OF SEEDS OF EACH TREATMENT IN EXPERIMENT 9, TRIAL 2

# a) <u>Differences between sowings</u>

Each cell in the sub-table consists of four rows which represent the four different temperature regimes included in the germination trial.

		Pumi od	SOWI Surface	MG 1 On-plant	Laboratory
$\Delta$ 1	P3	Buried S1=S2 S1=S2 S1=S2 S1=S2	\$1=\$2 \$1=\$2 \$1=\$2 \$1=\$2 \$1=\$2		S1=S2 S1=S2 S1=S2 S1=S2
SOWING 2 (Harvest	R1	S1=S2 S1=S2 S1=S2 S1=S2	\$1=\$2 \$1=\$2 \$1=\$2 \$1=\$2		S1=S2 S1=S2 S1=S2 S1\S2:20-24
	P1				S1=S3 S1=S3 S1=S3 S1=S3
M	P2	\$3\\\\$1:3-7 \$3\\\\\$1:8-10 \$1=\$3 \$1=\$3	S3\S1:2 S1=S3 S1=S3 S1=S3	\$1=\$3 \$1=\$3 \$1=\$3 \$1=\$3	\$1=\$3 \$1=\$3 \$1=\$3 \$1=\$3
SOWING (Harvest	. P3	\$1=\$3 \$1=\$3 \$1=\$3 \$1=\$3	S1=S3 S1=S3 S1=S3 S1=S3	S3\S1:7 S3\S1:8-10 S1=S3 S1=S3	S3(S1:7 S3(S1:8-10 S1=S3 S1=S3
	R1	S1=S3 S3§S1:10 S1=S3 S1=S3	S1=S3 S1=S3 S1=S3 S1=S3		S1=S3 S3\S1:8-10 S1=S3 S1=S3

# b) <u>Differences between harvests</u>

The following sub-table is arranged in a similar manner to the previous sub-table.

		Buried	HARVEST 2 Surface	Laboratory
	Pl	H10H2:3-7 H10H2:889 H1=H2 H1=H2	H10H2:2-7 H10H2:8%9 H1=H2 H1=H2	H1=H2 H1=H2 H1=H2 H1=H2
HARVEST 1	P2	H10H2:3-7 H10H2:8+10 H1=H2 H1=H2	H10H2:2-6 H10H2:9-13 H10H2:14 H1=H2	H1=H2 H1=H2 H1=H2 L1=H2
(Sowing 1)	P3	H10H2:4-7 H1=H2 H1=H2 H1=H2	H1(H2:2-7 H1=H2 H1=HC H1=H2	H2VH1:3 H1=H2 H1=H2 H1=H2
	R1	H10H2:7 H1=H2 H1=H2 H1=H2	H1(H2:1-7 H1(H2:8-10 H1=H2 H1=H2	H1=H2 H10H2:10-13 H10H2:14 H1=H2
HARVEST 1	P3			H1=H2 H1=H2 H1=H2 H1=H2
(Sowing 2)	R1		·	H1=H2 H1=H2 H1=H2 H1=H2
	P2	H2=H3 H2=H3 H2=H3 H2=H3	H2=H3 H2=H3 H2=H3 H2=H3	
HARVEST 3 (Sowing 3)	P3	H2=H3 H2=H3 H2=H3 H2=H3	H2=H3 H2=H3 H2=H3 H2=H3	
	R1	H2=H3 H2=H3 H2=H3 H2=H3	H2=H3 H2=H3 H2=H3 H2=H3	

### c) Differences between post-harvest treatments

## ) Collection P1 (Sowing 1 only)

H1	BURIED H2	Н3	H1 .	SURFACE H2
B=S B=S B=S	B=S B=S B=S B=S			
	BÖP: 3-7 BÖP: 8-13 BÖP: 14-19 BÖP: 20-24			SQP:3-7 SQP:8-13 SQP:14-1 SQP:20-2
BQL:2-7 BQL:8-13 BQL:14-19 BQL:20-24	B&L:3-7 B&L:8 B=L B=L		SQL:2-7 SQL:8-13 SQL:14-19 SQL:20-24	SQL:3-7 SQL:8 SQL:16-1 SQL:20-2

## ii) Collection P2

On-plant Surface

Lab.

		H1	BURIUD H2	Н3	H1	Н2
S1		S <b>§</b> B:2 B=S B=S B=S	S B: 4-7 S B: 8-10 B=S B=S			
IURLAGE	SZ		B=S B=S B=S B=S	B=S 3≬S:10&11 D=S B=S		
LINT	S1		B&P:6&7 B&P:8-13 B&P:14-19 B&P:20-24			SÕP: 4-7 SÕP: 8-13 SÕP: 14-1 SÕP: 20-2
ENTIALIO S3	S3		BQP:6&7 BQP:8-13 BQP:14-19 BQP:20-24	B=P B&P:8-13 B&P:14-19 B&P:20-24		SQP: 6&7 SQP: 8-13 SQP: 14-1 SQP: 20-2
Y210F	S1	BQL:1-7 BQL:8-13 BQL:14-19 BQL:20-24	BL:6&7 BL:8-13 BL:14-19 BL:20-24		S L:1-7 S L:8-13 S L:14-19 S L:20-24	SQL:4-7 SQL:8-13 SQL:14-1 SQL:20-2
LIBORITOIX	<b>S</b> 3		BQL:6-7 BQL:8-13 BQL:14-19 BQL:20-24			501:687 501:8-13 501:14-1 501:20-3

	SURFACE H2	II3	H1	ON-PLANT H2	Н3
				**************************************	
•	SQP:3-7 SQP:8-13 SQP:14-19 SQP:20-24				
	SQL:3-7 SQL:8 SQL:16-19 SQL:20-24			P=L P=L P=L P=L	

	н2	Н3	H1	H2	Н3
	SQP:4-7 SQP:8-13 SQP:14-19 SQP:20-24				
	SÖP: 6&7 SÖP: 8-13 SÖP: 14-19 S <b>ÜP: 20</b> -24	SQP:5-7 SQP:8-13 SQP:14-19 SQP:20-24			
<del></del>	SQL:4-7 SQL:8-13 SQL:14-19 SQL:20-24		•	P=L P=L P=L P=L	
	SQL:687 SQL:8-13 SQL:14-19 SQL:20-24			P=L P=L P=L P=L	

## iii) Collection P3

	H1	BURIED H2	Н3	H1	SURFACE H2
S1	S≬P:283 B=S B=S B=S	S S S S S S S S S			
SURFACE SO	5≬B:2-6 B=S B=S B=S				
চ ১১		B=S B=S B=S B=S	B=S B=S B§S:14-19 B§S:20-24		
S1		BÖP: 6&7 BÖP: 8-13 B=P B=P			S P: 6&7 S P: 8-13 S=P S=P
ON-PLANT					
S3		BÖP: 3-7 BÖP: 8-13 BÖP: 14-16 B=P	B=P B\(\Omega\text{P}:10-13\) B\(\Omega\text{P}:14-19\) B\(\Omega\text{P}:20-24\)		SQP:6&7 SQP:8-10 S=P S=P
31 S	BQL:4-7 BQL:8-13 BQL:14815 B=L	B=L B=L B=L B=L		S≬L:2-7 S≬L:8-10 S=L S-L	S=L S=L S=L S=L
LABORATORY SO N	BQL:7 BQL:8-13 BQL:14&15 B=L			SVL:2-7 SVL:8-13 SVL:14-17 S=L	
⊦i 53		B=L B=L B=L B=L			S=L S=L S=L S=L

SURTACE H2	<u>H3</u>	. Н1	ON-PLANT H2	田3
				·
P:6&7				
P:8-13 P				
P:6&7 P:8-10	S=P S=P			
P:6&7 P:8-10 P P	S=P S=P			
L L L			LOP:6-7 LOP:8-13 P=L	
r F			P=L P=L	
<u> </u>				
			L XP: 3-7	
L L L L			LQP:3-7 LQP:8-13 LQP:14-16 P=L	
L		_1		



## iv) Collection Rl

	H1	BURIED H2	. Н3	H1	SURFACE <u>H2</u>
s1	S≬B:1-6 B=S B=S B=S	F=S B=S B=S B=S			
SULLINGE	S\S:3 B=S B=S B=S				
S <b>3</b>		B=3 B=5 B=3 B=5	B=S B=S B=S B=S		
<i>.</i> S1		B&P:7 B&P:8-13 B&P:14&15 B=P			SQP:7 SQP:7-13 SQP:148:15 S=P
ON-PLANT					
S3		B=P B&P:10 B=P B=P	B=P B=P B=P B=P		S=P S=P S=P S=P
S1	B(L:7 B(L:8-13 B=L B=L	BQL:7 BQL:8-13 BQL:14&15 B=L		SQL:1-7 SQL:8-13 S=L S=L	SQL:7 SQL:8-13 SQL:14&15 S=L
LABOLATORY S	BQL:4-7 BQL:8-13 BQL:14-17 B=L			SQL:3-7 SQL:8-13 SQL:14-17 S=L	
S3					

URFICE H2	Н3	H1	ON-PLINT H2	нз
P:7 P:7-13 P:148:15 P				
				·
P P P	SIP:4&7 SIP:10-13 S=P S=P		·	
L:7 L:8-13 L:14&15 L			P=L P=L P=L P=L	
	<u></u>	<del></del>		

#### v) Collection H7 (Sowing one, Harvest two only)

	BURIED	SURFACE	OH-PLANT
SURFACE	B= S B= S B= S		
ON-PLANT	B≬P:4-7 B≬P:8-13 B≬P:14-17 B=P	SQP:4-7 BQP:8-13 SQP:14-17 S=P	
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#### d) Differences between collections

In the following sub-tables each block contains nine cells arranged in three columns and three rows. The columns represent sowings (from left to right, sowings one to three) and the rows represent harvests (from top to bottom, harvests one to three). The four rows within each cell represent the four temperature regimes included in the germination trial.

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## A5.2.3 Germination trial 3

### Statistical design

The statistical design of the comparisons analyzed for trial 3 was identical to that for trial 2 (see table A5.6) with the exception that seeds that had over-wintered in the field were not included. The comparisons that were analysed are described by the same numbers as the corresponding comparisons in trial 3.

The days on which observations of germination were made are described in the following pages as days 1 to 9. Table A5.16 provides a key for determining the actual day on which each observation was made.

TABLE A5.16

THE DATES ON WHICH OBSERVATIONS WERE MADE OF GERMINATION IN EXPERIMENT 9, TRIAL 3

Day number (as described in the following pages)	Date of observation	Number of days from the date of sowing
0 1 2 3 4 5 6 7 8 9	13th May 21st May 1st June 11th June 17th June 22nd June 28th June 2nd July 10th July 22nd July	8 19 29 35 40 46 51 59 71

# nalysis of germination totals

Germination totals (cumulative) were analysed for nine different days on which the number of newly emerged seedlings was recorded. Table A5.17 indicates the weights that were

used in attempts to stabilise the variance of each comparison. The weights were derived in two ways depending on which figure was used as the denominator in determining percentages of germination. When germination was expressed as a percentage of seeds sown, weights were computed from the numbers of seeds sown in each treatment averaged within each level of a particular factor (or factors). When germination was expressed as a percentage of the total number of seeds that ultimately germinated in that treatment, then weights were computed from the ultimate germination totals everaged within each level of a particular factor (or factors).

TABLE A5.17

WEIGHTS USED IN ATTEMPTS TO STABILISE THE VARIANCE OF COMPARISONS AMONG THE DATA OF EXPERIMENT 9, TRIAL 3

Compa- rison	Factor within which weights are proportional	Weights for each level of the appropriate factor
9/1 SS	В	Buried: 344, Surface: 155
TG	В	Buried: 344, Surface: 155
9/2 SS	C	Buried: 219, Surface: 170, On-plant: 500
ТG	В	P1: 144, P2: 197, P3: 82, R1: 344, H7: 48
	BC	Buried       Surface       On-plant         P1:       94       47       64         P2:       162       82       111         P3:       67       34       46         R1:       283       143       193         H7:       39       20       27

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Comp- rison	Factor within which weights are proportional	Weights for each level of the appropriate factor
9/3 SS	C	Buried: 250, Surface: 170, On-plant: 500
TG	AC	Buried       Surface       On-plant         P2:       288       144       172         P3:       144       72       86         R1       576       288       344
9 <b>/4 BS</b>	C	Buried: 149, Surface: 142, On-plant: 500
ТG	В	P2: 134, P3: 64, R1: 218
	BC	Buried       Surface       On-plant         P2:       188       126       243         P3:       86       58       112         R1:       294       197       380
9/5 SS	No weig	hts available.
TG	· Λ	P2: 80, P3: 40, R1: 140
	ABC	Harvest 2 Hervest 3  Buried Surface Buried Surface  P2: 346 194 108  P3: 173 97 97 54  R1: 606 339 339 189
9/6 SS	S A	P3: 282, R1: 211
T	i A	P3: 160, R1: 413

### Homogeneity of variance

The homogeneity of the variance of each comparison was determined by Bartlett's test. The test was performed on data both with and without weights. In those comparisons in which weighting gave a lower chi-square a weighted analysis of variance was subsequently performed. Otherwise, an unweighted analysis of variance was performed.

Table A5.18 presents the results of Bartlett's tests

(both weighted and unweighted) for each comparison.

## Analysis of variance

Table A5.19 presents the results of analyses of variance of data in each comparison, for each of the nine days on which observations were made.

## Duncan's multiple range tests

The results of Duncan's multiple range tests are presented as composite tables as described for trial 2 (see page 574). The comparisons chosen for inclusion are those indicated in table A5.14.

TABLE A5.18

THE RESULTS OF BARTLETT'S TESTS OF THE COMPARISONS AMONG THE GERMINATION DATA OF EXPERIMENT 9, TRIAL 3

Weighted anglysis Weights chi <sup>2</sup> SIG				ABC 25 * * ABC 24 * * ABC 26 *
Weighted englysis Weights chi <sup>2</sup> SIG	WB 25 ms WB 25 ms WB 25 ws WB 25 ws WB 25 ws WB 25 ws WB 22 ms WB 21 ms WB 21 ms	WB 25 **  WB 24 ns  WB 24 ns  WB 26 ns  WB 50 ns  WB 55 **  WB 75 **  WB 75 **  WB 75 **	70 78 78 78 79 79 79 79 79 79 79 79 79 79 79 79 79	77. 25 ** 77. 24 ** 77. 26 ** WA 26 **
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Note: Abbreviations are listed on page 30 .

TABLE A5.19

THE RESULTS OF ANALYSIS OF VARIANCE OF COMPARISONS AMONG THE GERMINATION DATA OF EXPERIMENT 9, TRIAL 3

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Note: Abbreviations are listed on page 556.



#### TABLE A5.20

THE RELATIONSHIPS BETWEEN MEAN PERCENTAGE GERMINATION OF SEEDS OF EACH TREATMENT IN EXPERIMENT 9, TRIAL 3

#### a) Comparisons between sowings

Each cell in the following sub-table consists of two rows which represent the two different expressions of germination. The upper row represents germination as a percentage of the seeds sown and the lower row represents germination as a percentage of the ultimate total germination.

		Buried	SOWING ONE Surface	On-plant
	P2			
SOWING TWO (Harvest one)	Р3	S1=S2 S1=S2	S1=S2 S1=S2	
	Rl	S1=S2 S1=S2	S1=S2 S1=S2	
	P2	\$1=\$3 \$1=\$3	S1=S3 S1=S3	S1=S3 S3\S1:1&2
SOWING THREE (Harvest two)	<b>P</b> 3	S1=S3 S1=S3	\$1=\$3 \$1=\$3	\$3\\$1:5-7 \$3\\$1:6-8
(	Rl	\$1=\$3 \$1=\$3	S\$\( \)\$1:1-9 \$1=\$3	\$30\$1:3-7 \$30\$1:3-7

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# b) Comparisons between harvests

		HAR' Buried	VEST 2 Surface
	Pl	H10H2:2-7 H10H2:5&6	H1=H2 H1∮H2:2-8
HARVEST 1	P2	Hl=H2 Hl=H2	H1≬H2:1-9 H1=H2
(Sowing 1)	<b>P</b> 3	H1=H2 H2(H1:3	H1(H2:1-5 H1(H2:1-8
	Rl	Hl=H2 Hl=H2	H10H2:1 H10H2:1
	P2	H2=H3 H2=H3	H2=H3 H2=H3
HARVEST 3 (Sowing 3)	<b>P</b> 3	H2=H3 H2=H3	H2=H3 H2=H3
•	Rl	H2=H3 H2=H3	H2=H3 H2=H3

(continued)

d) Companisons between port-harvest thesatments

Collection P4 (sowing one only)

			Γ						Γ					
HZ			H5				3≬P:1-9 S=p		135					
SURFACE H2		S=P S=P	SURFACE H2			SIP:1-9 SIP:1,2,4,8	S P: 1-9 S=P	SUZAKIOS	H2				3≬P:6-9 S=P	
FI			H1						디					
HZ			H3		B=S B[S:2-4		BAP: 1-9 BAP: 2-4		三五			B=S B <b>X</b> S:4-6		
BURIND	Bğs:1-7 Bğs:2-7	BMP: 1-9 BMP: 2-7	BURIND H2	B§S:1-9 B=S	318:1-9 B=8	ENP:1-9 BNP:1-7	BÅP:1-9 BÅP:1-4		EE EE	B08:2-5 B08:2-5		B 8:1-5	B 9 3 4 5 6 - 8 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	
, ,	B\(\begin{align*} \text{B\(\beta\):2-9}\\ \text{B\(\beta\):2-7}\\ B\(\beta\):2-7		Collection P2	SIB:1,3,5 B=S				Collection P3	H7	ы ш п п	B=S B=S			
	SURFACE	ON-PLANT	ii) <u>Co</u>	SITREACE	ν σ	Ñ	ON-PLANT S3	, (iii		S	SUKEACE	Ω KV	rs.	

E BC	B=S					
Ω N		303:1-5 303:2-5	B=S B§S:4-6			
ž		B			3≬P:6-9 S=P	
OM-PLANTS						
SO.		BIP: 1-5	307:1-9 807:2-5		S≬P:6-9 S=P	S=P P§S:4&5
iv) (vi	Collection R1	BURISD	H2	H1	SURPACE H2	H3
δ	B=S B=S	B=8 B=8				
SURFACE S2	B=S					
Ω		######################################	B=S B=S			
Ñ		D&P:1-9 B&P:1-8			SIP: 1-9 SIP: 1-7	
ON-PLANTS2						
Ω N		BIP:1-9	B&P:1-9		888:1-9 88:1-4	SQP: 1-6
<b>→</b>	Collection E	H7 (sowing one BURIED	only)	1	SURFACE	н
SURFACE	H7	H2 B=S	H2	H	717	
OM-PLAMP		B			S=P	

 $\widehat{\sigma}$ 

Seeds that wintered beneath the soil surface

Ω 22					P2175:1-9 P2175:1	P2 <b>\P3:</b> 1-9 P2 <b>\P</b> 5:1	83			
면 22 22							<u>H7</u> S2			
A. powellii				P2=P3 P2=P3	P21P3:1-9 P21P3:1		<u>ස</u>		P1=E7 P1=H7	
between collections of A. powellii							e species 83			
1							ns between the R1			
(1) <u>Comparisons</u>	P1=P2 P1=P2	P1 P2:5		P1=P3 P1=P3	P1=P3 P31P1:2-6		(2) Comparisons	P1=R1 P1=R1	R1 12-9 R1 17:2-6	
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F2&F3:1		Ω K													
		<u>形</u> 82													
		ν.	2		P1=H7 P1=H7			P2147:1-9 P2147:1-6			P3=H7 P3[H7:2-6				
	species	א ט						P2121:1-9 P2121:1	P2=R1 P2=R1		R1 (F5:5-7 R1 (F5:5	R1≬P3:1-9 P3=R1			-
	is between the	쩐 8	200							P3=R1 P3=R1					
	(2) Comparisons	2	בי	P1=R1 P1=R1	R1 X P1: 2-9 R1 X P1: 2-6		P2=R1 P2=R1	P2(R1:1-9 P2(R1:1		R11873:1-9 P3=R1	R1185:1,4-7 P3=R1			R1117:1-9 R1117:3-6	
H	ا ت		L	H	H ZH	I 口	<u>                                     </u>	ES ES	H	H	P2 H2	H	LI	ZH ZH	KH .

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1				•								
						Ω 20					P2187:6-9 P2183:1-6	P21P3:5-9 P2=P3
						지 전 전 전 전 전 전 전 전 전 전 전 2 2 2 2 2 2 2 2						
					of A. powellii	ζ.				P21P3:1-9 P2=P3	P2&P3:1-9 P2&P3:1-5	(
R118P3:1-9 P5=R1				soil surface	collections of	ਲ ਹ						
				aterd on the	between	집	30					
		R1 117:1-9 R1 17:3-6		Seeds that wintered	(1) Comparisons		P2 P1: 1-9 P2 P1: 2-7			P1=P3 P3[P1:2-7		
H	H	L CH	田	1. (ii			II	N FI	五 2		C	日
		ZE		•				C: P:			<b>P</b> 33	



	83								·				
717	32												
	31		P1 XH7:88.9 P1=H7			P2(147:1-9 P2(147:1-5			P3≬H7:5-9 P3=H7				
e species	83					P2=R1 P2=R1	P2=R1 R1≬P2:2-4		R1185:1-9 R1185:3,5&6	R11873:1-9 R11873:1-6			
ns between the R1	32							P5=R1 R11P3:2-5					
(E) Comparisons	ΣΩ.	R1 (P1:1-9 R1 (P1:2-7	R1 1 1 1 - 9 R1 1 1 1 : 2 - 7		P2=R1 P2=R1	P2=R1 P2=R1		P3=R1 P3=R1	P3=R1 P3=R1			2 R1 NT: 1-9 H7=R1	HZ
)		H	P7	H	H	P2 H2	EH H	H	P2 H2	五	H	EH ZH	H

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A5.2.4 Germination trial 4

TABLE A5.21

STATISTICAL DESIGN OF THE ANALYSES OF DATA FORMING EACH COMPARISON IN EXPERIMENT 9, TRIAL 3

Compa-	Factor code	Experimental factor	No. of levels	Levels	
9/11	В	Incubator shelf	2	Upper shelf, lower shelf	
	C	'other'	3	Harvest 1-Buried/buried, Harvest 2-Buried/buried, Harvest3-Surface/buried	
9/12	 A	Collections	3	P2, P3, R1	
	В	Post-harvest conditions	2	Buried/buried, Surface/buried	
	-C	Sowings	2	Sowing 1, sowing 2	
9/13	3	Collections	3	P2, P3, R1	
	C	Harvests	2	Harvest 1, Harvest 2	
9/14	В	Collections	2	P1, R1	
	C	Harvests	2	Harvest 1, Harvest 2	
9/15	B	Collections	2	P1, R1	
	C	Post-harvest treatments	; 2	Buried/buried, Surface/buried	
9/15	C	Collections	<u></u> 3	P1, R1, H7	
Mote:	Printed on mage 556.				

# A5.2.4.1 Analysis of viabilities

The number of seeds used in each replicate of each treatment was the same. Thus there was no possibility of using weights to reduce any heterogeneity among the variances.

TABLE A5.22A

THE RESULTS OF BARTLETT'S TEST AND ANALYSIS OF VARIANCE OF COMPARISONS AMONG THE VIABILITY DATA OF EXPERIMENT 9, TRIAL 4

BRROR	<b>ಕ</b>					 	
	2 24	3.40 5.61	0.73 ns	Φ			
AC ABC	N.	5.61	0.39 ns			1	
Variance ratios	~	4.26	46.4	~	5.32	· • • • • • • • • • • • • • • • • • • •	
riance AB	2	5.67	0.82 ns		١0	 	
Va	_	4.26	4.38	     <b>\</b>	5.32	4.1.4	
Д	۲-	4.26	3.94 ns		5.32	σ 7 *	
ধ	2	2.40	0,* 0,*	 		! ! ! ! !	i
Shi S	11	19.68	K) • •	2	7.81	7.1 ns	 
+ + 0	1	values: p=0.05 p=0.01	Observed SIG	DF Critical	values: p=0.05 p=0.01	Observed	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1
•	9/12			9/15		40.	

Note: Abbreviations are listed on page 556.

### Bartlett's test and the analysis of variance

Table A5.22A presents the results of Bartlett's tests and the analyses of variance for those comparisons which include different post-harvest treatments.

# Duncan's multiple range tests of the means

The analysis of differences in viability is only of interest in comparisons between post-harvest treatments. Thus table A5.22B presents results for these comparisons only.

TABLE A5.22B

THE RELATIONSHIP BETWEEN MEAN VIABILITIES OF SEEDS SUBJECTED TO DIFFERENT POST-HARVEST TREATMENTS

Collection	Incubator shelf	Sowing 1	Sowing 3
P1 P2 P3 R1 R1	L U U L	BB=SB BB=SB BB=SB BB=SB BB=SB	BB=SB BB=SB BB=SB
Note: Abbrevi	iations are listed	on pago 56.	

# A5.2.4.2 Analysis of germination totals

Each germination percentage was transformed to the arcsin scale before analyses were performed. The differences in viabilities between different treatments were not large enough to warrant the use of weights in the analysis of variance.

# Homogeneity of variances and analysis of variance

Table A5.23 presents the results of Bartlett's test and the analysis of variance for each comparison, for each day

TABLE A5.23

# THE RESULTS OF BARTLETT'S TEST AND ANALYSIS OF VARIANCE OF COMPARISONS AMONG THE GERMINATION DATA OF EXPERIMENT 9, TRIAL 4

ABC ERROR	7		24	
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os AC			2 2 2 5 6 7 6 9	2.00 0.00 0.00
e ratios BC	20 60 90 90 80 80 80	• • # 4 * • # • # • # • # • # • # • # • # • # •	4.26 7.82	.04 ns 0.7
Variance AB			2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2	0 • 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
C V	E∕ K		1 4.25	0.8 ns .04
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Ą			2 2 40 5.61	0.8 118 **5
chi <sup>2</sup>	711.07		19.68	7*7 11.2 12.3
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Comparison	11/6		9/12	

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ns ns ns ns ns ns ns ns ns ns ns ns ns n	40 . 44 . 45 . 45 . 45 . 45 . 45 . 45 .
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D 4 C * C * C * C * C * C * C * C * C * C	
* C H O * W * W * W * W * W * W * W * W * W *	7 7 7 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8 8
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	2/13

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DETY .	·	
70		
0.3 ns 0.4 ns 0.4 ns 0.8 ns	7.75 7.32 7.32 7.32 7.32 7.32 7.32 7.53 7.54 7.54 7.55 7.55 7.55 7.55 7.55 7.55	5.32 11.26
AB		9
0.0 0.3 0.3 0.3 0.8 0.8	7. 7. 4 dt * 5. * 6. * 6. * 6. * 6. * 6. * 6. * 6.	26 75 7 26 77 7 27 7 27 2
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Item Observed SIG Observed SIG Observed SIG	DF Critical values: P=0.05 p=0.07 Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observed SIG Observ	DF Critical Values: P=0.05
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Mote: Abbreviations are listed on page 556

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of the jormination trial.

#### Duncan's multiple range tests

The results of Duncen's multiple range tests of Mallarances between the means are presented in composite table 15.25. Table A5.24 indicates which composisons have been used to construct the composite table.

TABLE A5.24

A LIST OF DUPLICATE COMPARISONS BETWEEN FACTOR MEANS ALONG THE GERMINATION DATA OF EXPERIMENT 9, TRIAL 4

Harve ste	Sowing 1	Sowing 3
1	9/13 (BB ( <u>P2,P3,R1</u>	
	9/14 (BB P1,R1	
3	9/12 (BB,SB P2,P3,R1	9/12 ( <u>BB,SB</u> ( <u>P2,P7,R1</u>
	9/13 (BB (P2,P3,R1	
	9/14 (BB (P1,R1	
	9/15 (BB,SB (P1,R1	
	9/16 (BB ( <u>P1,R1,H7</u>	
Totas: 1	- The comparisons chosen for tuble 5.25 are underlined.	inclusion in

2 - Abbreviations are listed on page 556.

#### TABLE A5.25

THE RELATIONSHIPS BETWEEN MEAN PERCENTAGE GERMINATION OF SEEDS OF EACH TREATMENT IN EXPERIMENT 9, TRIAL 4

#### a) Comparisons between sowings (Harvest two only)

	P2	P3	Rl
Buried/buried	S1=S3	S1=S3	S1=S3
Surface/buried	S1=S3	S1=S3	S1=S3

#### b) Comparisons between harvests (Sowing one only)

Pl	P2	P3	R1		
H1=H2	H1=H2	H1=H2	H1=H2		

(All comparisons are for seeds from the Buried/buried treatment)

#### c) Comparisons between post-harvest treatments

	Pl	P2	P3	Rl
<b>\$1</b>	BB=SB	BB=SB	BB=SB	BB=SB
\$3		BB=SB	BB=SB	BB=S

(All comparisons are for seeds from Harvest two)

# d) Comparisons between collections

Each cell in the following sub-tables contains two rows. The upper row represents seeds from the "Buried/buried" treatment and the lower row represents seeds from the "Surface/buried" treatment. All possible comparisons can be made by considering the two sub-tables that follow.

,	HL E	<u>°3</u> H2	H1	<u>R1</u> H2
S1	P2¶P3:2-10	P2 P3:2-4 P2 P3:2-4	P2≬Rl:2	P2¶R1:2-4 P2=R1
<u>P2</u> S3		P2(P3:2 P2(P3:2		P2=R1 P2=R1

	<u> </u>	1 <u> </u>	H1.	<u>L</u> H2
S1 <u>R1</u>	R1≬P1:2-10	R1≬P1:2-19 R1=P1		
83				
S1		H7≬Pl:4-10		R1≬H7:2-3
<u>#7</u> \$3				

Note: Abbreviations are listed on page 56.

A5.3 Experiment 10

A5.3.1 Germination trial 1

#### Comparison 10/1

#### TABLE A5.26

STATISTICAL DESIGN OF THE ANALYSIS OF COMPARISON 10/1 FROM AMONG THE DATA OF EXPERIMENT 10, TRIAL 1

	Amperimental factor		Levels
Ċ	Position of inflorescence		Terminal, basal.
В	Other treat- ments	31	All other treatments for which a pair of values corresponding to factor C (above are available.

#### Bartlett's test and the analysis of variance

The results of Dartlett's tests and the analyses of variance are presented in table A5.28. All data were transformed from percentages to arcsins before analysis.

#### Comparison 10/2

#### TABLE A5.27

STATISTICAL DESIGN OF THE ANALYSIS OF COMPARISON 10/2 FROM AMONG THE DATA OF EXPERIMENT 10, TRIAL 1

Factor code	Experimental factor	No. of levels	Levels
A	Collections	7	P1, P3, P5, R1, R4, R5, H7
В	Position of inflorescence	2	Terminal, basal
C	State of utricle	2	Present, absent

# Bartlett's tests and analyses of variance

Table A5.28 presents the results of Bartlett's tests and the enalyses of variance. For ten of the days of the

effect and significant interaction. For these days, the analyses were repeated and the replicates were pooled for that factor which showed no effect and no interaction.

Those revised analyses of variance also are presented in table A5.28. All data were transformed from percentages to arcsins before the analyses were performed.

#### Duncan's multiple range test of the means

Table A5.29 presents the results of Duncan's multiple range tests of the means. This table is composed of subtables which portray comparisons of the means of each level of the different factors examined. In each sub-table four rows within each cell represent the four temperature regimes included in the germination trial.

TABLE A5.28

THE RESULTS OF BARTLETT'S TEST AND ANALYSIS OF VARIANCE OF COMPARISONS 10/1 TO 10/3 AMONG THE DATA OF EXPERIMENT 10, TRIAL 1

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Companiaon		10/2 (revised analyses)

Hote: Abbreviations are listed on page 556.

#### TABLE A5.29

THE RELATIONSHIPS BETWEEN MEAN PERCENTAGE GERMINATION OF SEEDS OF EACH TREATMENT IN COMPARISON 10/2

The four rows within each cell of the following sub-tables represent the four sequential temperature regimes included in the germination trial.

# a) Comparisons between seeds with and without utricles

	Terminal	Basal
P1	+U=-U +U=-U +U=-U +U\$-U:22&23	+U=-U +U=-U +U=-U +U\$-U:22
P3	+U=-U +U=-U +U0-U:17-19 +U0-U:20	+U=-U +U0-U:11&12 +U0-U:14-19 +U0-U:20
P5	+U=-U +U=-U +U=-U +U=-U	+U=-U +U=-U +U=-U +U=-U
Rl	+U=-U +U=-U +U=-U +U=-U	+U=-U +U=-U +U=-U +U=-U
R4	+U=-U +U=-U +U0-U:17-19 +U0-U:20	+U==U +U=-U +U0-U:17-19 +U0-U:20
R5	+U==U +U==U +U==U +U==U	+U=-U +U=-U +U=-U +U=-U
н7	+U=-U +U=-U +U=-U +U=-U	+U=-U +U=-U +U=-U +U=-U

## b) Comparisons between seeds from different positions

	+Utricle	-Utricle
Pl	Bot: 6&7 Bot: 8 B=T B=T	Bot: 6&7 Bot: 8 B=T B=T
P3	T§B:6 B=T B=T B§T:21	B=T B=T TØB:14 B=T
P5	B=T B=T B=T B=T	B=T B=T B=T B=T
Rl	B=T B=T B=T B=T	B=T B=T B=T B=T
R4	B=T B=T B=T B=T	B=T B=T B=T B=T
R5	B=T B•T:9 B=T B=T	B=T B@T:9 B=T B=T
н7	B=T B=T B=T B=T	B=T B=T B=T B=T

(continued)

## c) Comparisons between collections and species

## i) Between collections of A. powellii

	P	1	P3						
	+0	<b>-</b> U	+0	_U					
т Р3	Pl=P3 Pl(P3:9-13 Pl=P3 Pl=P3	P1=P3 P1@P3:10-13 P1@P3:14 P1=P3							
В	P1=P3 P1\P3:8-11 P1=P3 P1=P3	P1=P3 P1\P3:10-12 P1=P3 P1=P3							
T	Pl=P5 Pl(P5:9-13 Pl(P5:14-19 Pl(P5:20-25	P1=P5 P1(P5:10-13 P1(P5:14-19 P1(P5:20-25		P3=P5 P3(P5:13 P3(P5:14-19 P3(P5:20-25					
P5 B	P1=P5 P10P5:9-13 P10P5:14-19 P10P5:20-25	P1=P5 P10P5:8-13 P10P5:14-19 P10P5:20-25		P3=P5 P3(P5:13 P3(P5:14-19 P3(P5:20-25					

## ii) Between collections of A. retroflexus

	R		R4 +π <b>−</b> U						
	+0	-U	+0						
Ŧ	R1=R4 R1=R4 R1=R4 R1=R4	R1=R4 R1=R4 R1&R4:16-19 R1&R4:20&21							
R4 B	R1=R4 R1=R4 R1@R4:16-19 R1@R4:20&21	R1=R4 R1=R4 R1@R4:16-19 R1@R4:20&21							
T	R1=R5 R50R1:10-13 R50R1:14-19 R50R1:20&21	R1=R5 R5@R1:10-13 R5@R1:14-19 R5@R1:20&21	R4=R5 R5@R4:10-13 R5@R4:14-19 R5@R4:20	R4=R5 R5@R4:10-13 R5@R4:14-19 R5@R4:20&21					
R5 B	R1=R5 R5@R1:10-13 R5@R1:14-19 R5@R1:20	R1=R5 R50R1:10-13 R50R1:14-19 R50R1:20	R4=R5 R5@R4:10-13 R5@R4:14-19 R5@R4:20&21	R4=R5 R5@R4:10-13 R5@R4:14-19 R5@R4:20&21					

	-19 -25	-19 -25	.19 .25	27.0	.19 .25	<u> </u>	
<u>P5</u> U	P5=R1 P5=R1 R1≬P5:16- R1≬P5:20-	P5=R1 P5=R1 R1≬P5:16- R1≬P5:20-	P5=R4 P5=R4 R4\P5:17- R4\P5:20-	₽5=R4 ₽5=R4 R4≬P5:18& R4≬P5:20-	P5=R5 R5(P5:13 R5(P5:14- R5(P5:20-	P5=35 R5105:10- R5195:14- R51P5:20-	75-H7
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<u>P2</u> _U	P3=R1 P3&R1:11-13 P3&R1:14-16 P3=R1	₽5=¤1 ₽5=¤1 ₽5≬¤1:16-17 ₽5=¤1	F3=84 F3134:11-13 F3184:14-19 F3184:20 21	₽5=34 ₽5≬R4:15 ₽5≬R4:14-19 ₽5≬R4:20821	P2=R5 P3=R5 R5\P3:16-19 P3=R5	P5=R5 R51P3:8-15 R51P3:14-17 P3=R5	11 4CF
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#### Comparison 10/3

This analysis was performed in two stages, firstly using data for seeds from terminal inflorescences and secondly using the data for seeds from basel inflorescences. The design of the analysis was identical for both stages; it involved a hierarchical analysis of variance.

TABLE A5.30

STATISTICAL DESIGN OF THE ANALYSIS OF COMPARISON 10/3 FROM AMONG THE DATA OF EXPERIMENT 10, TRIAL 1

<u> Level</u>	Experimental <u>level</u>	Number of categories	Contribution of categories to the next level
0	Replicates	72	3 to each category of level 1
1	Plants	24	(3,3,2,1,3,3,3,3) to level 2 categories respectively
2	Collections	9	(1,3,5) to level 3 categories respectively
3	Species	3: <u>A.</u>	hybridus, <u>A. retroflexus</u> and powellii

## Homogeneity of variance and analysis of variance

Table A5.31 presents the results of Bartlett's test and the hierarchical analysis of variance.

## Revised comparisons

Each of the two comparisons that were initially analysed were sub-divided into A. powellii and A. retroflexus components. The samples of A. hybridus were omitted. Table A5.32 presents the results of the revised hierarchical analyses of variance. The design of the new analyses contained one less level than the original analyses.

## Duncan's multiple range tests of the means

range toots between means of the highest level of each enalysis. The blocks within each sub-table consist of two cells, the upler cell representing terminal seeds, the lower cell representing basal seeds. The four rows within each cell represent genuination at the different temperature regimes included in the governation trial.

TABLE A5.31

THE RESULTS OF BARTLETT'S TESTS AND THE HIERARCHICAL ANALYSIS OF VARIANCE OF COMPARISON 10/3, AMONG THE DATA OF EXPERIMENT 10, TRIAL 1

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Mote: Ibbrevietions are listed on page 556.

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HIERARCHICAL NG THE DATA	7 DEN	9		19				2 6	2 6	20	10	9	200	2 <u>C</u>		10	9	9	125	2 :	2 5	5 5	9	9	29	2,5	9	2	99	9	2
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TESTS AND COMPARISON)	T & C	41	ባሬ	) [~	-ယ	ON.	6;	<u>-</u> (	7 7 7	/ ナ	- ረ- - ቢ/	1,6	[~(	<b>か</b> (	0 V	- 8	101	25		ĽΛ	ഗ	<b>~</b> 00	റ	0,7	<u></u> ,	<u>, , , , , , , , , , , , , , , , , , , </u>	7-7-	. L . ħ	<u>7</u> 7	-0	20
THE RESULTS OF BARTLETT'S		A. powellii																	A. powellii												
THE RESULT		compartson terminal	seed																basal	3000 C	} } }										

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	retroflexus	retroflexus
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	terminal seed	្ឋ

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Level 2 VR DFD DFN SIG	(	* N 0.	12 2 01	6.93 11 2 *	4 20	2 14	2 25 2	0 TX 2	2 349	
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Day	7	ភ វ៉	<u> </u>	200	20	21	22	U) V	CI C	
Comparison Species										

Mote: Abbreviations are listed on page 556

#### TABLE A5.33

# THE RELATIONSHIP BETWEEN MEAN PERCENTAGE GERMINATION OF SEEDS OF EACH TREATMENT IN COMPARISON 10/3

The four rows within each cell of the following subtables represent the four sequential temperature regimes included in the germination trial.

#### a) Comparisons between collections of A. powellii

	Pl	P2	P3	P5
T P2	P1=P2 P1(P2:9-13 P1(P2:14-19 P1(P2:20-25			
В	P1=P2 P1 P2:10-13 P1 P2:14-19 P1 P2:20-25			
T P3	Pl=P3 Pl&P3:9-13 Pl&P3:14-16 Pl=P3	P2=P3 P2=P3 P3 P2:15-19 P3 P2:20-25		
В	Pl=P3 Pl(P3:10-13 Pl(P3:14-16 Pl=P3	P2=P3 P3 P2:10-13 P3 P2:14-19 P3 P2:20-25		
T	Pl=P5 Pl(P5:10-13 Pl(P5:14-19 Pl(P5:20-25	P2=P5 P2=P5 P2=P5 P2=P5	P3=P5 P5@P3:9 P3@P5:15-19 P3@P5:20-25	
P5 B	P1=P5 P10P5:10-13 P10P5:14-19 P10P5:20-25	P2=P5 P2=P5 P2=P5 P2=P5	P3=P5 P3 P5:10-13 P3 P5:14-19 P3 P5:20-25	
T	Pl=P6 Pl&P6:9-13 Pl&P6:14-17 Pl=P6	P2=P6 P2=P6 P6 P2:16-19 P6 P2:20-25	P3=P6 P3=P6 P3=P6 P3=P6	P5=P6 P5 P6:9 P6 P5:16-19 P6 P5:20-25
P6 B	Pl=P6 Pl0P6:9-13 Pl0P6:14-19 Pl0P6:20-25	P2=P6 P2=P6 P6 P2:15-19 P6 P2:20-25	P3=P6 P3 P6:9-13 P3 P6:14-19 P3 P6:20-25	P5=P6 P5=P6 P60P5:16-19 P60P5:20-25

## b) Comparisons between collections of A. retroflexus

		R1	R4
R4	T	R1=R4 R1=R4 R10R4:16-19 R10R4:20&21	
	В	Rl=R4 Rl=R4 Rl@R4:16-19 Rl@R4:20&21	
R5	T	R1=R5 R5(R1:10-13 R5(R1:14-17 R1=R5	R4=R5 R5@R4:10-13 R5@R4:14-19 R5@R4:20&21
ny	В	R1=R5 R5(R1:10-13 R5(R1:14-17 R1=R5	R4=R5 R50R4:10-13 R50R4:14-19 R50R4:20&21

Note: Abbreviations are listed on page 556

#### Comparison 10/4

TABLE A5.34

# STATISTICAL DESIGN OF THE ANALYSIS OF COMPARISON 10/4 FROM AMONG THE DATA OF EXPERIMENT 10, TRIAL 1

	Factor code	Experimental factors		Levels
10/4 <u>A</u>	A	Collections	6	H7, R1, R4, R5, P3, P5
	В	State of the utricle	2	Present, absent
	C	Light regime	2	Alternating light and darkness, continuous darkness
10/43	В	Collections	9	H7, R1, R4, R5, P1, P2, P3, P5, P6
	O	Light regime	2	Alternating light and darkness, continuous darkness

## Bartlett's tests and the analyses of variance

Table A5.35 presents the results of Bartlett's tests and the inclyses of variance. All data were transformed from percentages to arcsins before analysis.

## Duncan's multiple range tests

The results of Duncan's multiple range tests are presented in table A5.36. Each sub-table of table A5.36 consists of cells within blocks. The four rows within each cell represent comparisons of the germination after 13, 19 and 25 days (from top to bottom).

TABLE A5.25

THE RESULTS OF BARTLETT'S TEST AND ANALYSIS OF VARIANCE OF COMPARISON 10/4 AMONG THE DATA OF EXPERIMENT 10, TRIAL 1

TABLE A5.36

THE RELATIONSHIPS BETWEEN MEAN PERCENTAGE GERMINATION OF SEEDS OF EACH TREATMENT IN COMPARISON 10/4

The four rows within each cell of the following sub-tables represent, from top to bottom respectively, the germination after 13, 19 and 25 days.

_				·	· · · · · · · · · · · · · · · · · · ·	Ta Ta	į	
А								
P6								
ы								
А								
면							61 64 64 60 67 67	
H							# 100 m	İ
А					XXXX HHZ HHZ HHZ HHZ	X P S P S P S P S P S P S P S P S P S P	MP6 MP6 MP6	
P3					でまま	でませる	日立立	
H					2200 2000 2000 2000 2000 2000	222 222 222 222 222	2176 6175 3=76	
					P4 P4 P4	P1 P1 P1	७ ७ ७ ७	
А			2000 2000 2000 2000 2000 2000 2000 200		22=P5 22=P5 22=P5		42 = 54 54 \$2 50 45 50 50 50 50 50 50 50 50 50 50 50 50 50 50 50 50 5	
24			000 6464		NNN HHH		P6 1	
H			PSXPP PSXPP PSXPP		75=7 75=7 75=7 75=7		1000 1000 1000 1000 1000 1000 1000 100	
А	222		EEN		=    X   H   H   H   H   S   H   H		X P6 P60 P60 P60	
되	₽1±₽2 ₽1≠₽2 ₽1≬₽2		1200 1200 1200 1200 1200 1200 1200 1200		무무		현현현	
H1	222 1425 1425 1425 1425 1425 1425 1425 1		200 200 200 200 200 200 200 200 200 200		**************************************		1 PG 1=PG 1=PG	
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		(기 단		57		F F		Н Р

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		21≬P6 P5=21 R1≬P6		P6=R4 P6≬R4 P6=R4		351P6 P6=R5 P6=R5	· ·	H7≬P6 P6=H7 P6=H7	
		R1=F6 R1&P6 R1&P6		P6=R4 P6≬R4 R4≬P6		RSNP6 RSNP6 RSNP6	-	H7&P6 H7&P6 H7&P6	
+		P5=R1 P5=R1 R1≬P5	P5=R1 P5=R1 R1 &P5	P5=R4 P5=R4 R41P5	R4&P5 P5=R4 R4&P5	P5=35 P5=35 R5\R5		P5=H7 P5=H7 H7≬P5	₽5=E7 ₽5=H7 H7≬₽5
\		P5=R1 R1	R1	P5=R4 R4&P5 R4&P5	₽5=34 24≬₽5 24≬₽5	E5000000000000000000000000000000000000		H7	P5±H7 H7≬P5 H7≬P5
		P5=21 P5&21 P5=21	P	F3&34 P3&34 P3&84	₽5=24 ₽5=34 ₽3≬24	P5%25 P5%25 P5%85		520 520 520 520 520 520 520 520 520 520	P5MH7 P5MH7 P5=H7
0.7=0.7		P50001 R1005 R1005	P3=R1 P3=R1 P3=R1	P5084 P5084 P40P5	P3=R4 P3\R4 P3=R4	P3 R35 R5 R75 R5 R75	·	P5	P5=H7 P5=H7 P5=H7
ZINOI		P2=R1 P2=R1 R1		P2=R4 P2=R4 R4≬P2		P2=R5 P2=R5 R5&P2		P2=H7 P2=H7 H7≬P2	
2 <b>4</b> 195		P2=R1 R1		P2=R4 R4\P2 R4\P2		R		P2=H7 E7&P2 H7&P2	
OANLA		P1=R1 P1=R1 P1=R1		P1=R4 F1=R4 P1()R4		P1=R5 P1 = R5 P1 = R5		P1=E7 P1=H7 P1 (	
. 94=14		₽1 <b>≬R</b> 1 ₽1=R1 R1≬P1		P1		P10R5 R50P1 R50P1		17 8 TH 17 8 TH 17 8 TH	
<b>-</b>	Щ	<u>[</u>	Д	EI	Щ	EH	Щ	EH	щ

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# b) Comparisons between collections of A. retroflexus and between species

		L	Rl D	R L	4 D	R5 L D			
R4	T	Rl=R4 Rl R4 Rl=R4	Rl=R4 Rl=R4 Rl R4						
	В	R1=R4 R1∮R4 R1=R4	R1=R4 R1=R4 R1=R4						
ne.	T	R5¶R1 R1=R5 R1=R5	Rl=R5 Rl=R5 Rl@R5	R5≬R4 R5≬R4 R4=R5	R4=R5 R4=R5 R5≬R4				
R5	В								
н7	T	R1=H7 R1=H7 R1=H7	R1=H7 R1=H7 R1=H7·	H70R4 H70R4 R4=H7	R4=H7 R4=H7 H7≬R4	R5¶H7 R5=H7 R5=H7	R5=H7 R5=H7 R5=H7		
	В	R1=H7 R1=H7 R1=H7	R14H7 R1=H7 R1=H7	R4=H7 H7§R4 R4=H7	R4≬H7 R4=H7 H7≬R4				

	TI7	L=D L=D L=D	AAA HHH HHH
	R5		
	P.4-	L=D L=D L=D	11
between light regimes	Щ <b>.</b>	H KOKKI H H CA H H H	L=D L&D L=D
	P6	LLE BOD BOD	
	P5	111 111 111	L=D L=D
	P3	LXD L=D L=D	LL LX = D = D
on lich	2 2	L=D L=D L=D	
- 1	H	DAG    XX    HH	
Jornpari son	•	EI	Д
ा			

Note: Abbreviations are listed on page 556

A5.3.2 Germination trial 2

TABLE A5.37

# STATISTICAL DESIGN OF THE ANALYSIS OF THE DATA OF EXPERIMENT 10, TRIAL 2

Lovel	Emporimental level	Number of categories	Contributions of categories to the next level
0	Replicatus	40	4 to each category of lavel 1
1	Collections	10	(1,3,6) to level 2 categories respectively
2	Species	3: A. A.	hybridus, A. retroflexus and powellii

## Bartlett's test and the analysis of variance

Table 45.38 presents the results of Bartlett's tests and the hierarchical analysis of variance. All of the data were transformed from percentages to arcsine before analysis.

TABLE A5.38

RESULTS OF BARTLETT'S TEST AND THE HIERARCHICAL ANALYSIS OF VARIANCE OF THE DATA OF EXPERIMENT 10, TRIAL 2

D	2	SIG	VR	C DFN	LEVE colloc DFD	L 1 tions CIG	AīS	LEVEL Speci DEM		SIG
Day 9 250 360 561 751	chi 28.4 32.4 32.4 16.1 18.5 10.7 34.8	**  **  **  ns  ns  **  ns  ns	1.51 1.77 1.01 5.58 2.79 1.28 0.53	777775557	35000000000000000000000000000000000000	ns ns ns ** ** **	7.000 to the total of the total	0.000.000.000.000.00	1978 1139113780 113780	ns ns ns ** ** 25 25

Mote: Abbreviations are listed on page 556.

## Duncan's multiple range test of the means

Table A5.39 presents the results of Duncan's multiple range test of the means of level two, the species.

#### TABLE A5.39

THE RELATIONSHIPS BETWEEN SPECIES MEANS AS DETERMINED BY DUNCAN'S MULTIPLE RANGE TEST OF THE DATA FROM EXPERIMENT 10, TRIAL 2

		A. hybridus	A. powellii
Α.	powellii	PNH:40-61	
Α.	retroflexus	R=H	P)R:40-61

#### \5.4Dxperiment 11

#### TABLE A5.40

## STATISTICAL DESIGN OF THE ANALYSIS DATA FROM EXPERIMENT 11

Factor code	Experimental factor	No. of levels	Level
В	Soil type	7	BES, EEL, BCL, BG, HC, ML, ISL (See table 7.1% for key to abbreviations).
C	Collection	3	H7, P3, R1

## Bartlett's test and the analysis of variance

Table A5.41 presents the results of Bartlett's test and the analysis of variance. All of the data were transformed from percentages to arcsins before the analysis was made.

# Duncan's multiple range tests of the means

Table A5.42 presents a summary of the results of Duncan's multiple range tests of the means of each treatment for each day on which observations of germination were made.

TABLE A5.41

THE RESULTS OF BARTLETT'S TEST AND ANALYSIS OF VARIANCE OF THE GERMINATION DATA OF EXPERIMENT 11

ERROR	09																		
REPS	Ю	2.76	1.7 ns	1. 5. tr	2.75 ns	2.2 ns	۲. u	7. 18.	0 5 8	2.0	0.5	ns 0.4	rn	и • в С	0.3	0.2	Ø	ns.	
ABC																			
ratios 3 AC																			
Varience rat AB BC	12	20°0°	₩ %*	κ/* Θ.*	00 * 0.*	0 •* 0*	* ° *	w* •*	O	, , , , , , , , , , , , , , , , , , ,	11s 1 - 2	75 70 10 10	Ø	ວ F ທ	0	1.0	ns	• Ω	
Vari AB		·																	
೮	2	4.98	ス で * ・ な ・ な	104.1	50°4 4.	C**	76.4	₩ ₩ •	64.5	46.4	42.8	3.4 **	) <b>*</b>	×*	スト マン・マン・ハン・フィン・フィン・フィン・フィン・フィン・フィン・フィン・フィン・フィン・フィ	34.9		· ^*	
ല	9	01 kg 20 Kg	(1) * (4) * (4) *	43.4	* 27 * 5.3	% % * 70 * 7	0. € €	40 *	24°8 **	ν. ω. ω.	20.0	√π * ∞ ∞		 N *	01 00 * 01 *	29.5	* (		page <b>5</b> 56
ব																			цo
chi <sup>2</sup>	50	31.4	512.4	432.4	× 27.2.	N-77*	780	129.0	\$. 0. *	27.4	ns 24.1	22 . 21 <b>.</b> 4		X) K	101 102 103 103 103 103 103 103 103 103 103 103	ns 22.6	ពន	ns.	s are listed
Item	DF Critical	20.	Observed	Observed	Observed STG	Observed STG	Observed STG	Observed STG	Observed STG	Observed	Slu Observed	SIG Observed	SIG	Observed STG	Obscrved	Sign	on the contract of the contra	Observed SIG	Abbreviations
Day			~	<b>C</b> 1	К	7	77	Ø	2	ω	<u>ن</u>	9		<u>~</u>	0 <u>1</u> 7	13		4.	1:0te:

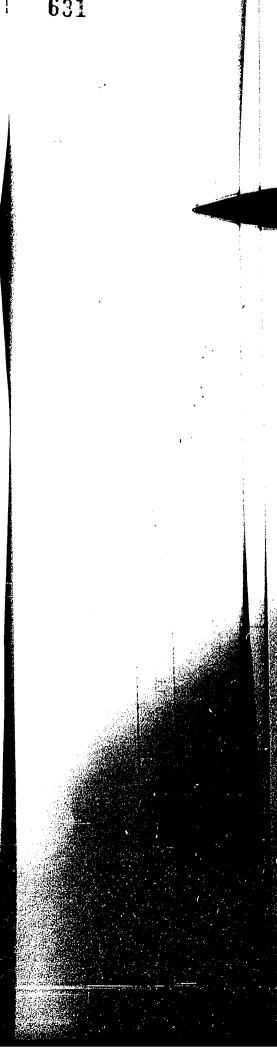


TABLE A5.42

THE RELATIONSHIPS BETYEEN MEAN PERCENTAGE GERLINATION OF SEEDS OF EACH TREATMENT IN EXPERIMENT 11

	<b></b>			1			1			<b></b>			:
PSL												·	
HC													POLÁHO: 5-78
BG										BGMHC:3-14	BG≬IIC:4-14	BG=HC	BG=PSL
BCL							B℃L=BG	BCL=BG	BCL=BG	BCL MIC: 3-14	BCL   HC: 8-14.	BCL=HC	BCL PSL:8-13
BEL				BEL BCL: 1-78	BEL BCL: 3-14	BKL≬BCL:6-14	BEL & BG: 1-14	BEL≬BG: 3-14	BEL≬BG:6,7,	BELŽIIC: 1-14	BELL IIC: 3-14.	BIID≬HC:6-14	BEL PSL:1-14
BES	BEL≬BES:1-14	BEL BES: 3-14	BEL BES: 6,7,	BES=BCL	BES=BCL	BES=BCL	BES=BG	BES=BG	BES=BG	BESMIC: 3-14	BESIIC:8-14	BES=HC	BES=PSL
	<u>P3</u>	RET	ZH	P2	k BGI	<u>ZE</u>	5 <sub>4</sub>	BG EG	<u> </u>	53	阿	HZ	P3

					1	,
P2	BES=PSL	BIL≬PSL:1-14	BCL≬PSL:8-13	BG=PSL	POL 180: 5-78	
FA FSD FSD	BES=PSL	BEL PSL: 3-14	BCL=PSL	BG=PSL	PSL \( \) HC: 8-14	
ZH	BES=PSD	BULNESL:6,78 9-14	BGT=EST	BG=PSL	IIC=PSI	
P2	正 § BIS:1-14	BELŽIL: 1&2	II \ BCL: 1-7%	III.≬BG:1-14	NI, MHC: 1-14	MIÅPSL:1-14
IM Fil	II \ BES: 3-14	BEL=M	11-14 III\BGL:3-14	III≬BG:3-14	III≬HC:3-14	MINEST: 3-14
HZ	ILNBES:6,7,14 BEL-ML	BEL=ML	BCL=ML	BG=LL	IIL≬HC:6-14	III & PSL:7,
						+ 10

# b) Comparisons between species

	1,						
R18H7	R1&H7:4,7	01 <b>−2∶</b> 7±§r፰	R1=H7	81≬H7:4-7	R1=H7	7H=FR	R1 & H7: 3-14
下285日7	₽5≬∃7:3-14	₽5≬H7:1-14	P3&H7:5-14	P5&H7:2-14	P3=H7	P3&II7:5-14	P5≬H7:1-14
P3&R1	P5≬21:6-8	₽ <b>5</b> ≬Ω1:1,2	P3(R1:5-14	P31R1:2	P3=R1	P3\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\	P5021:1,2,7,
•	BES	BEE	BCL	BG	DII	E CO	且

Note: Abbreviations are listed on page 556

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## A5.5 Experiment 12

## A5.5.1 Germination trial 2

## TABLE A5.43

# STATISTICAL DESIGN OF THE ANALYSIS OF DATA OF EXPERIMENT 12, TRIAL 2

Level	Experimental level	Number of categories	Contribution of categories to the next level
•	Replicates	124	4 to each category of level 1
1	Plants	31	(4,4,5,3,3,2,3,2,4,1) to level 2 categories respectively
2	Collections	10	(6,3,1) to level 3 categories respectively
3	Species: A. p	owellii, A.	retroflexus, A. hybridus

#### Bartlett's test and the analysis of variance

Table A5.44 presents the results of Bartlett's test and analysis of variance for each day of the germination trial. The data were transformed from percentages to arcsins before the analysis was made.

TABLE A5.44

# RESULTS OF BARTLETT'S TEST AND THE HIERARCHICAL ANALYSIS OF VARIANCE OF THE DATA OF EXPERIMENT 12, TRIAL 2

Day	chi <sup>2</sup>	SIG	<b>V</b> R	LEVI Plan DFD	EL 1 ats DFN	SIG	VR C	LEVI 11e		ns SIG	<u>v</u> r		EL §	SIG
1234567	92.3 414 367 293 241 207 183	**	242 2.2 0.9 2.7 6.3 14	93 93 93 93 93 93 93	21 21 21 21 21 21	** ** ** ** **	3.5 1.6 36 17 11 7.0 6.9	21 45 98 40 28 24 25	7 7 7 7 7	* ns ** ** ** **	0.1 .06 0.2 0.1 0.8 3.9 9.2	14 25 8 9 10	2222222	ns ns ns ns ns ns
8 9 10	185 158 159	**	8.9 7.3 7.0	93 93 93	21 21 21	**	6.8 7.1 7.4	26 27 27	7 7 7	** **	14 19 22	10 10 10	2 2 2	**

Note: Abbreviations are listed on page 556.

## Duncan's multiple range test of the means

Table A5.45 presents the results of Duncan's multiple range test of the means of level 3, i.e. the differences between species.

#### TABLE A5.45

THE RELATIONSHIPS BETWEEN MEAN PERCENTAGE GERMINATION OF SEEDS OF EACH TREATMENT IN EXPERIMENT 12, TRIAL 2

		A. hybridus	A. powellii
A.	powellii	H≬P;8-10	
A.	retroflexus	H=R	<b>R≬P:6-1</b> 0

Note: Abbreviations are listed on page 56.

#### A5.5.2 Germination trial 3

TABLE A5.46

STATISTICAL DESIGN OF THE ANALYSIS OF DATA OF EXPERIMENT 12, TRIAL 3

Factor code	Experimental factor	No. of levels		Levels
В	Collections	5	P1, P2, P3,	R1, H7
C	Temperature regimes	2	30°/15°C,	35°/20°C
Note:	Abbreviation	s are 1:	isted on pag	<sub>e</sub> 556.

## Bartlett's test and the analysis of variance

Table A5.47 presents the results of Bartlett's test and the analysis of variance. All data were transformed from percentages to arcsins before the analysis was made.

TABLE A5.47

THE RESULTS OF BARTLETT'S TEST AND ANALYSIS OF VARIANCE AMONG THE DATA OF EXPERIMENT 12, TRIAL 3

Day	Item	chi <sup>2</sup>	Va B	riance C	ratio BC	s ERROR
304,7	DF Critical values:	9	<u>+</u>	1	4	20
	p=0.05 p=0.01	16.9 21.7		4.35 9.10		
1	Observed SIG	29.0 **	4.57 **	0.01 ns	0.4 ns	
2	Cbserved SIG	76.5 **	0.6 ns	2.0 ns	0.8 ns	
3	Observed SIG	72.3 **	10.4	23 <b>.</b> 4 **		
4	Observed SIG	64.0 **	10.9	29.3 **	5.8 **	
5	Observed SIG	49.7 **	12.0 **	ジ5•3 **	7.4 **	
6	Observed SIG	34•2 **	14.7	65 <b>.</b> 7	9.0 **	
7	Observed SIG	32.4 **	18.2 **	92.3 **	9.9	
8	Observed SIG	33·3 **	21.0	97.2 **	2.9 ∗*	

The following values are after 8 days at 35/20 and 15 days at 30/15 and 35/20.

Observed SIG	14.7 ns	**	ns	<b>5.</b> 65
	•	**	ns	**

Mote: Abbreviations are listed on page 556.

## Duncan's multiple range test of the means

Table A5.48 presents the results of Duncan's multiple range test of the means.

#### TABLE A5.48

THE RELATIONSHIPS BETWEEN MEAN PERCENTAGE GERMINATION OF SEEDS OF EACH TREATMENT IN EXPERIMENT 12, TRIAL 3

**a)** Comparisons between temperatures

The left column in the following sub-table compares germination during eight days at 30°/15°C with germination during eight days at 35°/20°C and the right column compares germination after 8 days at 35°/20°C with germination after 15 days of which the first eight were at 30°/15°C and the remainder at 35°/20°C.

Pl	35≬30:3-8	35(8)=30-35(15)
P2	30=35	35(8)=30-35(15)
P3	35≬30:8	35(8)=30-35(15)
Rl	35≬30:6–8	35(8)=30-35(15)
H7	35≬30:3-8	35(8)=30-35(15)

## b) Comparisons between collections and species

The three rows within each cell in the following sub-table represent, from top to bottom respectively, 1 to 3 days at 35 /20 °C, 1 to 8 days at 30 /15 °C, and after 15 days (first 8 days at 30 /15 °C, the remainder at 35 °/20 °C).

	P1	P2	P3	Rl
Pl	P1\P2:3-8 P1=P2 P1=P2			
P3	P1 P3:3-8 P1=P3 P1=P3	P2=P3 P2=P3 P2=P3		
R1.	Pl@Rl:3-5 Pl=Rl Rl@Pl	R1≬P2:5-8 P2=R1 R1≬P2	R1≬P3:5-8 P3=R1 R1≬P3	
н7	H7≬Pl:5-8 Pl=H7 H7≬Pl	H7≬P2:3-8 P2=H7 H7≬P2	H7≬P3:3-8 P3=H7 H7≬P3	H7¶R1:3-7 R1=H7 R1=H7
Note:		ns are listed	on page 556.	

## A5.6 Experiment 13

TABLE A5.49

# STATISTICAL DESIGN OF THE ANALYSIS OF DATA OF GERMINATION TRIALS 2 TO 6 OF EXPERIMENT 13

Trial	Factor code	Experimental factor	No. of levels	Levels
12-12-68	¥	Wintering position	4	60cm above soil; 15cm above soil surface; 15cm below soil
	2	Collections	2	P2, P3
	C	Remains of inflorescence	2 2	with remains; without remains
21-1-69	A.	Wintering position	2	60 cm above soil; 15cm above soil
	В	Collections	2.	P2, 23
	C	Remains of inflorescence	2 e	with remains; without rormins
18-3-69	A.	Wintering position	3	60cm above; 15cm above; soil surface; 30 cm below; 15cm below
	В	Collections	2	P2, P3
	C	Remains of inflorescence	2 e .	with remains; without remains
3-5 <b>-</b> 69		Wintering position	5	60cm below; 15cm below
	3	Collections	5	P2, P3
	O	Remains of inflorescence	2 e	with remains; without remains

# Bartlett's tests and the analyses of variance

Table A5.50 presents the results of Bertlett's tests and the analyses of variance. All of the data were transformed from percentages to arcsins before the analyses were made.

TABLE A5.50

THE RESULTS OF BARTLETT'S TEST AND ANALYSIS OF VARIANCE OF THE GERMINATION DATA OF EXPERIMENT 13, TRIALS 2-6

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Note: Abbreviations are listed on page 556.

## Duncan's multiple range tosts of the means

Table A5.51 presents the results of Duncan's multiple range tests of the means.

## TABLE A5.51

THE RELATIONSHIPS BETWEEN MEAN PERCENTAGE GERMINATION OF SEEDS OF EACH TREATMENT IN EXPERIMENT 13, TRIALS 2 TO 6

This table is arenged as a series of sub-tables which begin on the following page.

Comparisons between seeds that had wintered with and without plant remains

3rd Lay				<b>-</b> R=+R	-RØ+R:4-11				-R=+R	-R=+R
18th April	-R≬+R: 3-12	-RX+R:8-12	-R=+R	-R≬+R:3-12	-R=+R	–3≬+R:2	-R=+R	-2=+R	-R=+R	-R=+R
18th Karch	-R=+R	-光=+유	꿈+=끕-			-R=+R	-R=+2	-		
21st Jen.	-R1+R: 3-12	-R=+R				<u>-R=+R</u>	-R=+R			
12th Dec.	-R=+R	-R=+R	-R=+R	- R=+R		-R=+R	-E§+R:4-10	-R=+B	-R≬+3:10-12	
	+60cm	+15cB	P2 00cm	-15cm	# <b>2</b> 0⊝−	#209+	+15cm	P5 00cm	-15cm	<b>−</b> 20cm

Comparisons between collections

7		12th Dec.	21st Jan.	18th Larch	18th April	Srd Tay
+R P2=P3 -R P2=P3	P2=P3 P2=P3		P2=P3 P21P3:324	P2=P3 P2=P3	P2=P3 P2≬P3:5-12	
+R P2=P3 -R P31P2:4-14	1		P2=P <b>3</b> P2=P <b>3</b>	72=7 72=75	P2=P3 P2≬P3:7-12	
+R F2=F3 -R F2=P3	I n.n.			€ <b>द=</b> टत € <b>द=</b> टत	₽3≬₽2:4-12 ₽3≬₽2:8-12	,
+R P2=P3 -R P2=P3	P2=P3 P2=P3				P2=P3 P2≬P3:5-11	P3\P2:6-11 P3\P2:6-11
#R R					P2=P3 P2=P3	F3\P2:5-11 P2=P3

## Comparisons between wintering positions

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In the following sub-table, the five rows within each cell represent, from top The different wintering positions are indicated in the body of the table by the codes shown within to bottom respectively, germinstion trials 2 to 5. parentheses at the row and column headings.

			<b> </b>	<u>i</u>	
15cm below (4)					
15( +)					
surface (3) -R			_		3=4
Soil su					z=4. •
15cm above (2) +R -R		,	2=3 2=3 213:7-12	285:4-14 2=5 382:7-12	2=4
15сп а +R			0 · 00 · 11 · 11 · 11 · 11 · 11 · 11 ·	2=3 502:4-12	2=4
above (1)	~~~. ~~~~ ~~~~~	7	1 = 2 1 = 2 1 = 3 2 = 2 2 - 1 2	183:138:14 1=3 381:4-12	7=4
60cm ab +ஐ	ľ	7777 0000	L . L . H II	7=5 7=5 5 <u>0</u> 7:4-12	7=4
	2	above (2)	P2 Soil	surface (3)	

	4=5 5 <u>0</u> 4:485	• • • + + + + + + + + + + + + + + + + +		 200. 4.5.4	512:7-12	•••(;; ••	511:5-12		below (5) P3
	• • • • • • • • • • • • • • • • • • •	4=5 4\$5:28:3	513:2-12	5=5	• • • ¢; •	• • • ፡ ፡ ፡ ፡ . ቢ	••• <u>~</u> •	· • • <u>/</u> •	59 mo09
· ·			5=4 • 5≬4:586 •	3=4· • 3=4·	214:4-9 • 2=4	2=4 • • \$2:5-12	7=	1=4 • •¥1:5-12	below (4) P3
		·	>=4 403:2-12	0 • • 6 • 6 • 6 • 6 • 6 • 6 • 6 • 6 • 6	7=4 • - 7 - 7 • 4	2=4 • ?=4	7=4 7=4	7=4· · 7=4· ·	강.
	·				2≡3 3≬2:7-12	2=3 5 <u>0</u> 2:4-12	1=5 3 <u>8</u> 1:4-12	7=5 5 <u>8</u> 7:4-12	r) P)
					213:4-14	S=2	113:13:14	1=3	surface (5)
					212:7-12	10 ·	115:2-12	- T •	P2   Soil

Note: Abbrevictions are listed on page 556.