FACTORS INVOLVED IN THE INCIDENCE OF POTATO SKIN SPOT AND IN INFECTION BY THE CAUSAL ORGANISM *OOSPORA PUSTULANS*

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SUMMARY

1. An examination of factors at lifting and in storage in relation to skin spot development in a field crop of King Edward potatoes showed that time of haulm destruction had no effect on the disease development, but if tubers were lifted early and stored in boxes then a reasonable level of control of the disease could be achieved. In later lifted tubers and tubers stored in clamps for a few weeks after lifting, skin spot could only be effectively reduced by organo-mercury disinfection, the limit of the period when the treatment was effective coinciding with the maximum depth of penetration of the fungus into the tissues. Alternative non-mercury fungicides tested proved ineffective in skin spot control.

2. Organo-mercury disinfection treatments of seed tubers reduced disease in the resulting crop, but if carried out at planting time tended to delay plant emergence. Treatments where seed was boxed at lifting showed better emergence than that from seed which was clamped.

3. Microscopic examination of the eyes of tubers in storage showed that a progressive spread of infection appeared to occur under humid and cool conditions, but this was not, in the main, attributable to secondary infection during storage. Under conditions favourable to disease development a large proportion of infected eyes were killed in the 12-16 week period after lifting.

4. In an attempt to simulate the effects of rainfall over the

lifting period, by the application of water to the soil, it was found that moderate rather than high and low levels of soil moisture favoured skin spot infection.

5. Field and small scale studies showed that low temperatures, especially over the lifting period and in the first few weeks of storage, were associated with a high level of skin spot development.

6. It was demonstrated that, for several varieties, planting seed with a high disease inoculum will result in a high level of skin spot infection of the resulting crop. It was also shown that the adverse effects of delaying plant emergence and blanking from planting heavily infected seed could be reduced by growing in light soils or using vigorously sprouting varieties. There was some evidence that skin spot infection was less favoured on sandy soils than on medium-loam soils.

7. Oospora pustulans was found to grow better on a sugar rather than a starch medium and to be able to break down polysaccharide molecules to monosaccharide units, thus the high sugar contents found in susceptible varieties subjected to conditions associated with high skin spot development may encourage infection.

INTRODUCTION

Historical

The earliest record of what was probably potato skin spot came from Scotland in the middle of the 19th century when Johnston (1845) described a condition in potatoes known as "pock and blind eye". The symptoms occurred in early spring when the tuber surface was covered with small round spots or pimples and eyes thus infected did not sprout. The disease was attributed to an insect. This information was quoted by Moore (1951).

Carruthers (1904) gave a fuller description of the disease, describing the tubers as being covered with numerous bluish-black warts of about ¼ in. diameter. When thinly peeled a dark brown patch was found below each wart. On dissecting these patches the cells of the potato were found to be permeated by threads of very fine mycelium. The causal organism was not, however, identified.

The name skin spot was first ascribed to the disease by Pethybridge (1915) but the fungus in the infected tissue was identified as Spicaria solani. Several subsequent workers tried unsuccessfully to identify the causal organism suggesting Spicaria nivea (Milburn and Bessey, 1915), Rhizoctonia crocorum (O'Brien, 1919) and Phoma eupyrena (Wollenweber, 1920). Gussow (1918) attributed the disease to a physiological disorder brought about by badly ventilated storage conditions particularly when the potatoes were covered

with soil.

Owen (1919) was the first worker to make a thorough investigation of skin spot and identified the fungus isolated from the infected tissues as Oospora pustulans (Owen and Wakefield). The fungus was grown in pure culture, fully described morphologically and classified systematically. Owen was unsuccessful, however, in inoculation experiments with the fungus attempting to reproduce skin spot symptoms on potato tubers.

Shapovalov (1923) in America claimed that there was no conclusive proof that Oospora pustulans or any other of the suggested fungi was the primary cause of the disease, claiming that the pustules of skin spot, being very similar to the closed and immature sori of powdery scab, were merely the early stages of this disease. However, Millard and Burr (1923) cleared up the controversy by successfully carrying out inoculation experiments with Oospora pustulans and Spongospora subterranea proving that skin spot and powdery scab were distinct and that Oospora pustulans was responsible for skin spot.

Geographical Distribution

Skin spot appears to be confined to the temperate and cooler regions of the world; besides being present in Britain it has been reported from Norway, New Zealand, Tasmania, Canada (Anon, 1950), Germany (Fuchs, 1954), U.S.S.R. (Hrobruih, 1953), Sweden (Emilsson, 1960) and U.S.A. (Folsom and Bonde, 1950). This is in accordance with the fact that the optimum growth temperature of the fungus is relatively low (Owen, 1919;

Fuchs, 1954; Salt, 1957; Kharkova, 1961b).

Economic Effects

Skin spot infection in normal circumstances is a superficial attack restricted to the outer layers of cells of potato tubers. In the past it has been of little economic significance with ware tubers since skin spot pustules are easily removed by peeling. Nowadays, however, with an increasing market for washed and pre-packed ware tubers, skin spot infection is of more significance since the appearance of the potato tubers is an important selling factor.

There have been exceptional cases where abnormally deep skin spot lesions have resulted in ware crops being unsaleable. This happened in a crop treated with a sprout depressant isopropyl phenyl carbamate, I.P.C., (Ives, 1955), and it has also occurred in crops left to overwinter in the ground (Boyd and Lennard, 1962).

The main economic effect of skin spot lies in eye infection. Infected eye tissue can be so severely damaged that the buds will either fail to sprout at all or there will be a considerable delay in sprouting. It has been demonstrated with the variety King Edward that, where tubers showing varying levels of skin spot infection are planted, there is a delay in emergence corresponding to the degree of eye infection. Such delays decrease the length of the growing season for the individual plants concerned and so decrease the yield thereof. In the most extreme cases of eye infection,

where all eyes are severely infected, a considerable level of blanking occurs.

Total crop yields, except in extreme cases, are not greatly affected by the adverse effects on individual plants of blanking and delay in emergence because of the compensating growth of neighbouring plants, more especially if the crop is close planted. There is evidence, however, that skin spot infection can reduce the proportion of seed to ware and also decrease tuber numbers. This is of concern economically in a crop grown for seed (Boyd, 1957; Boyd and Lennard, 1961b).

Oospora pustulans will infect potato roots and stolons and this has been connected with a reduction in tuber numbers (Salt, 1958). Reductions in final yield of plants grown from seed of varying levels of skin spot have been reported, but this has been variable from season to season (Hirst et al., 1965, 1966, 1967).

Planting of infected seed will result in a spread of the disease to the resulting crop. The contrast between the amount of disease on produce from heavily infected seed and on that from seed free from visible infection or with low levels of infection has been very marked (Boyd and Lennard, 1961a; Edie, 1964).

Symptoms

The external symptoms of skin spot do not become evident until at least two months after infection and in Britain under normal field conditions recognisable pustules are not fully

developed until February or March (Allen, 1957; Boyd and Lennard, 1961b; Nagdy, 1962). The typical symptoms which do occur are in the form of small distinct black or purplish-grey pimples on the tuber surface of 0.5 - 2.0 mm. in diameter extending in depth to rarely more than 2.0 mm. (Millard and Burr, 1923) and not normally associated with any general rotting of the tuber tissue. The pustules may occur singly or in aggregated groups covering a large area of the tuber.

The form of the pustules can be variable. Owen (1919) considered that the skin texture of the tuber variety had an effect on the pustule type varying from a pimple in the coarse skinned Arran Chief tubers to a small sunken lesion with a slightly raised centre in a smooth skinned variety such as King Edward. Pustules of an intermediate type were found on other varieties.

Millard and Burr (1923) observed that crater-like depressions occurred in older pustules on King Edward tubers.

Kharkova (1961a) classified the disease symptoms into the categories of superficial flat lesions, prominent roundish pustules often with pressed edges, depressed lesions of irregular form and deeply depressed roundish little holes, and attributed these differences to two forms of the fungus and to different races of these forms. Lesions similar to the latter two described by Kharkova were reported in Britain in tubers treated with the sprout depressant I.P.C. (Ives, 1955) and tubers overwintered in the ground (Boyd and Lennard, 1961b).

Environmental conditions during development of infection

have an important effect on the form and size of the skin spot pustules (Todd, 1963).

The effect of Oospora pustulans on the potato plant is not confined to the tubers. It has been demonstrated that the fungus isolated from brown lesions in the cortex of badly damaged root systems of Majestic potatoes was identical to Oospora pustulans and that Oospora pustulans isolated from infected tubers was pathogenic of root systems of potato and tomato plants (Hirst and Salt, 1959).

The Causal Organism

The causal organism of skin spot is Oospora pustulans (Owen and Wakefield), a member of the Hyphomycetales (Fungi Imperfecti).

The mycelium consists of narrow septate filaments 2-4 μ in diameter, hyaline or pale brown in colour. Conidiophores of length up to 260 μ are numerous and there is repeated and irregular branching from them. The chains of conidia break up easily producing individual conidia which are either oval-cylindrical or cylindrical single-celled structures with rounded ends 6-12 μ by 2.0 - 2.5 μ (Owen, 1919; Kharkova, 1961a). Dark coloured sclerotia ranging from 49 μ to 1 mm. in diameter have been demonstrated (Kharkova, 1961a; Lennard and Boyd, 1965; Hirst, Hide and Stedman, 1965).

Oospora pustulans will grow well on a wide range of media, but is best on a cooked vegetable medium. Growth on uncooked vegetable tissue only occurred on potato of the tissues

investigated and was much slower. This suggests that the fungus can exist saprophytically on dead organic matter in the soil displaying a slight parasitism to potato tubers (Owen, 1919).

Kharkova (1961b) maintains that Oospora pustulans grows best on media containing sugar and related this to the fact that conditions where there were high sugar contents in the tuber could be associated with high skin spot infection. Biochemical changes have been observed within infected tubers including the breakdown of starch to sugar (Gomolyako, 1959; Vartapetyan, 1962).

The fungus has an optimum growth temperature at around 15°C. Growth virtually ceases at temperatures as low as 0°C and as high as 24°C (Owen, 1919; Salt, 1957; Kharkova, 1961b). It has also been reported that cultures of the fungus could survive temperature extremes of -39°C during winter and 45°C for up to 4 hours (Kharkova, 1961b). There are no records of survival limits of the microsclerotia produced in culture.

Growth appears to be best within the pH range of 6.2 - 6.8, but it ceases at the extremes of pH 4 and pH 10 (Kharkova, 1961b).

Infection of the Tuber

(a) Infection process

The result of infection by Oospora pustulans is the production of skin spot pustules on the surface and in the eyes of tubers, sometimes completely killing or severely damaging the buds.

Within mature pustules the cells are seen to be completely disorganised and starch grains have entirely disappeared. Many of the cells are dark brown with thick, cuticularised walls and interspersed with them are clear cells with hyaline walls. Septate hyphae of the fungus can be found in quantity in the pustules. A well marked, cup-shaped layer of cork, often six or more cells deep, surrounds the pustule, extending on either side to within a few cells of the outer periderm isolating the infected region from the healthy tissue below (Owen, 1919; Millard and Burr, 1923). The importance of the cork barrier in this respect is emphasised where the sprout depressant I.P.C. was applied to tubers (Ives, 1955) and where tubers overwintered in the ground (Boyd and Lennard, 1961b). In both of these cases abnormally deep lesions were formed and no cork barrier was found to be present.

Allen (1957) investigated the infection process and suggested that in infected areas, as large as mature pustules, hyphae could be detected and cell walls had thickened 8 days after inoculation and a cork barrier was established after 16 days. Nagdy (1962) disputed this, suggesting that while hyphae could be detected in infected tissue after one month, the effects of cell wall thickening and cork barrier establishment were not in evidence until two months after inoculation. Both found that obvious skin spot symptoms were not visible until two months after inoculation and Nagdy (1962) stated that mature pustules took 3-4 months to form.

(b) Paths of infection

It has been suggested that Oospora pustulans penetrates the tuber through parts of the surface beneath which there is no cork barrier, i.e. lenticels, buds themselves and the area between the buds and the cork periderm (Greeves and Muskett, 1939; Allen, 1957).

It is well established that wounds and abrasions of the periderm allow entry of the fungus (Fuchs, 1954; Boyd and Lennard, 1961a; Nagdy, 1962).

Nagdy (1962) has evidence that penetration can be made through an intact periderm but this is not conclusive.

Kharkova (1961a) suggests that infection occurs through eye tissue since there is no cork layer beneath it, that it occurs through lesions of powdery and common scab and that the fungus can penetrate through stolons to infect new tubers.

(c) Time of infection

By disinfection experiments, Greeves and Muskett (1939) suggested that skin spot infection occurred at or some time before normal lifting time. No reduction in skin spot was brought about by lifting the crop early. Boyd (1957) demonstrated that tubers lifted in early September developed substantially less skin spot than those lifted at the normal time in early October. Allen (1957) showed that tubers were more susceptible to skin spot when they were mature but Nagdy and Boyd (1965) disagreed with this.

Hirst, Salt and Hide (1963) have shown that there is a

build up of Oospora pustulans in the soil during the period of tuber formation and this reaches a peak value in late September thus there is a greater supply of fungal inoculum at normal lifting time as opposed to an earlier lifting time.

Kharkova (1961a) suggested that infection takes place in the field, but that there is a secondary infection during storage. Edie (1966) demonstrated a progressive increase of Oospora pustulans found on tuber eyes and in the number of eyes which were killed with the fungus present, on tubers during damp storage.

Persistence and Transmission

It is well established that the most important source of infection for the transmission of skin spot comes from planting infected seed, the higher the level of seed inoculum the higher will be the infection of the resulting crop irrespective of environmental conditions (Boyd and Lennard, 1961a; Edie, 1964; Hirst, Hide and Stedman, 1966).

Regarding infection from the soil Owen (1919) demonstrated that Oospora pustulans could grow well under saprophytic conditions suggesting that the fungus could survive on dead organic matter in the soil.

It has been suggested that development of skin spot on tubers grown from apparently symptomless seed tubers is evidence of Oospora pustulans occurring naturally in some soils (Anon, 1939; Boyd and Lennard, 1961a), but the demonstration that fungal infection can be detected on so-called

skin spot free seed by incubation and microscopic examination (Hirst and Salt, 1959) renders the above assumption invalid.

Using the indicator plants of rooted potato stem cuttings or tomato seedlings, the presence of the fungus in certain soils which had not carried a crop of potatoes for periods ranging up to 10 years, has been reported (Hirst and Salt, 1956; Salt, 1957; Nagdy, 1962). Kharkova (1961a) maintained that infected underground plant parts could overwinter and still produce viable conidia the following year.

Soil Factors

It was reported (Anon, 1932) that certain soils were more suited to the development of skin spot than others. In Russia it has been suggested that infection is most severe on a sandy-podzol soil (Khrobrykh, 1959) and that there was greater infection on a ferruginous podzol than peat boggy soil (Gomolyako, 1959). In Britain the development of a method of investigating soil infectivity (Hirst, Salt and Hide, 1963) has resulted in the examination of various soil types showing that stem base infection was least on neutral peat and alluvial soils, more on light loams and most on clay soils (Salt, 1964).

Climatic Factors

The importance of climatic factors on skin spot development has been established by examination of temperature and rainfall data over a period of 37 years revealing a connection between above average rainfall at lifting time, lower than

average temperatures during the first three months of storage and severe skin spot outbreaks (Boyd and Lennard, 1962). Another factor which must be taken into consideration in accounting for seasonal variation in skin spot, is the level of the disease on the planted seed. A high level of seed inoculum tends to increase infection in the resulting crop. Since 1958 detailed examination of the three factors has been made and it has been shown that in years when there has been a high incidence of skin spot at least one of the factors has been satisfied (Boyd, McGee and Lennard, 1966).

The reports that infection is favoured by humid conditions both before and after inoculation of tubers (Allen, 1957) and that low temperature and high humidity during storage result in a high level of skin spot development (Edie, 1964) are in agreement with the general climatic observations.

The effects of skin spot of delay in emergence and blanking can be aggravated by weather conditions in early spring which result in a cold, dry soil (Boyd and Lennard, 1961b).

Varietal Factors

There are marked differences in the susceptibilities of potato varieties to skin spot and they can be grouped according to degree of susceptibility. Of the more commonly grown varieties Golden Wonder and Dunbar Rover are the most resistant while Kerrs Pink, Craigs Royal and King Edward are highly susceptible. Varieties like Home Guard and Arran Pilot are

about midway between the two extremes (Anon, 1932; Boyd, 1954, 1957; Boyd and Lennard, 1961a; Nagdy and Boyd, 1965).

There is evidence that varieties with a thicker periderm and higher crude fibre content in the skin tended to be more resistant to skin spot infection indicating possibly a greater mechanical resistance to fungal penetration (Nagdy and Boyd, 1965).

In the comparison of varieties, according to infection from artificial inoculation with Oospora pustulans, it has been shown that there is a highly significant correlation between surface and eye infection (Nagdy and Boyd, 1965), but in the comparison of varieties, according to natural infection, the susceptibility to surface and eye infection may not necessarily be related (Anon, 1932; Boyd, 1957; Boyd and Lennard, 1961a). It appears that, in the case of natural infection, variable conditions of humidity and temperature in the field may be such as to encourage eye infection more than surface infection or vice versa (Nagdy and Boyd, 1965).

The variety King Edward has been found to be particularly susceptible to eye infection resulting in blanking and irregular emergence in crops (Boyd, 1954, 1957; Boyd and Lennard, 1961a).

The fact that a variety is highly susceptible to skin spot is no criterion that when badly infected seed is planted there will be serious delays in emergence or extensive blanking. Basic varietal characteristics may be able to overcome the harmful effects of skin spot. An example of

this can be seen when tubers of the varieties King Edward and Arran Pilot show equally severe eye infection. In the field, however, the eventual loss in plant establishment may be quite different (Boyd, 1957). There is little doubt that the difference is due to sprout vigour (Nagdy and Boyd, 1965).

Varieties appear to react differently to tuber infection and to stem-base infection since the order of skin and eye susceptibility bears little resemblance to the order of susceptibility of stem-bases as given by Salt (1964).

Control

The most efficient method of control of skin spot is by disinfecting tubers with an organo-mercury solution at lifting and subsequent box storage time. Any delay in treatment reduces the effectiveness of the control measure (Greeves and Muskett, 1939; Foister, 1943; Boyd, 1957, 1960; Edie and Boyd, 1966). While controlling skin spot development on the seed there is also evidence that this treatment will reduce the infection of the resulting crop to some extent (Edie, 1964).

Washing tubers to remove surface soil has been reported to reduce skin spot if carried out at lifting time and box storage (Boyd, 1957).

A considerable level of control of skin spot in comparison to what will develop in clamp storage can be brought about by boxing tubers at lifting (Boyd, 1957, 1960; Boyd and Lennard, 1961a; Edie and Boyd, 1966).

Boyd (1957) demonstrated that lifting tubers one month before normal lifting time resulted in a considerable reduction

in skin spot. This is not in agreement with Greeves and Muskett (1939) who found that early lifting gave no control measure at all.

Removal of haulm one month before normal lifting was found to give some control of skin spot (Boyd, 1957).

According to Edie (1964) disinfection of seed with organo-mercury solution at planting does reduce skin spot in the resulting crop and appears to have no phytotoxic effect on the sprouts in terms of delay in emergence or yield decrease.

Application of non-mercury fungicides at planting was found to give a reduction in Oospora pustulans infection of the root parts (Salt, 1958).

Introduction to Experimental Work

The experimental work was designed to gain more knowledge of the factors involved in determining the incidence and severity of skin spot on potato tubers and of the effects of the disease on a growing crop and relating this to possible cultural methods of control of the disease or its economic effects.

The work may be considered in the following four sections.

A. Factors at lifting and in storage in relation to skin spot development, with particular reference to the effects of level of soil moisture and temperature and humidity conditions in storage.

B. Seed tuber treatments for the control of skin spot in relation to subsequent field effects and skin spot

development in the following crop.

C. Field responses to varying levels of skin spot infection on seed tubers of different varieties planted in different types of soil.

D. Carbohydrate nutrition of Oospora pustulans and its relationship to carbohydrate changes in potato tubers during storage.

GENERAL MATERIALS AND METHOD

Since skin spot is mainly a problem of seed potatoes the tubers used for experimental work were usually of a seed size grade, i.e. tubers passing through a $2\frac{1}{4}$ in. mesh but not through a $1\frac{1}{4}$ in. mesh riddle. In most cases this was left to visual judgement. In a large part of the experimental work the variety used was King Edward, as this commercially important variety is highly susceptible to skin spot and the associated delay in emergence and blanking (Boyd, 1957; Anon, 1963).

1. Assessment of surface infection

Tubers to be assessed were first of all washed and a standard method of assessment (Boyd, 1957) was used based on the following categories as determined by visual judgement.

Severe (S) - $\frac{1}{4}$ or more of the surface area affected.

Moderate (M) - between $\frac{1}{10}$ and $\frac{1}{4}$ of the surface area affected.

Slight (L) - from trace up to $\frac{1}{10}$ of the surface area affected.

Trace (T) - up to 10 pustules.

A surface infection index (S.I.I.) for any given sample of potatoes can be calculated by multiplying the number of tubers in each category by the average percentage area affected:

Severe x 62.5

Moderate x 17.5

Slight x 5

Trace x 1

This total is then divided by the total number of tubers examined (N) and multiplied by 100/62.5 to give a mean percentage area affected. Thus the surface infection index is given by the formula

$$S.I.I. = 1.6\frac{(62.5 \times S + 17.5 \times M + S \times L + T)}{N}$$

As in any form of visual assessment the method is subject to personal bias on the part of the assessor, but it is reasonably well standardised to give a consistent rating of samples by different workers. It may be noted, however, that the weighting of 62.5 for the severe disease category in the above formula is probably excessive. This figure is derived from the middle of the range 25 to 100 per cent surface area affected, but in practice few tubers in this category have a percentage area affected much above 25 per cent.

2. Assessment of eye infection

2.1. Visual examination

An eye was considered to be infected if there was a skin spot pustule within the eye tissue. In some cases the bud tissue may be completely killed, while in others the sprout may be developed, although the growth would probably be impaired.

Three categories of tuber infection were used:

All eyes infected.

Some eyes infected.

No eyes infected.

An eye infection index (E.I.I.) was calculated by multiplying the number of tubers in the 'all eyes infected' category (A) by 100 and the number in the 'some eyes infected' category by 50, dividing the sum of the products by the total number of tubers examined (N) and multiplying the resulting figure by 100 to obtain a mean percentage of eyes infected (Nagdy, 1962). The formula for eye infection index may thus be expressed as

$$E.I.I. = \frac{(100 \times A + 50 \times S)}{N} \times 100$$

This method has one serious drawback, in the assumption that in the 'some eyes infected' category the average percentage of eyes infected will be 50 per cent. However, in a severely infected sample, most eyes of tubers in the 'some eyes infected' category will tend to be infected, whereas in a slightly infected sample few eyes of tubers in this category will be infected. Thus severely infected samples may tend to be underestimated and slightly infected samples will tend to be overestimated. A solution to this problem would appear to be in bringing in other categories of eye infection, but this would involve more time in assessments. Some measure of eye infection is, however, necessary since this is of such economic importance.

2.2. Microscopic examination

This technique demonstrates that tuber eyes may be infected with Oospora pustulans although there are no visible signs of skin spot pustules or any obvious eye damage. It involves excising tuber eyes and incubating in a humid atmosphere at 18°C. This treatment produces aerial conidiophores of the fungus easily identified under the microscope (Salt, 1957).

The procedure followed was to thoroughly wash the tubers to be examined in running water to remove surface contamination. The eyes were then excised using a $\frac{3}{8}$ in. diameter cork borer with a fitted spring loaded plunger to eject the plug of tissue. The plunger had a concave end to reduce damage to the tuber eye and it ensured that a standard depth of the plug was removed each time. The irregular end of the plug was removed with a scalpel. When eyes were taken from the rose end of the tubers there were often more than one eye on a plug. When this situation occurred each eye was assessed separately.

In the work described here tuber samples were normally approximately 40 tubers and 50 eyes were randomly selected for incubation. Eyes which were greened or blighted were discarded. Tests were made comparing the figures obtained using varying numbers of eyes from a sample and it was found that 50 eyes gave similar results to larger numbers of eyes.

The excised plugs were set up on damp blotting paper in plastic boxes with moist cotton wool attached to the inner lid of the box. This ensured a humid atmosphere. Incubation

was carried out at 18°C for 5 days in darkness and then the plugs were examined under a dissecting microscope. The eyes were classified into 4 categories:

Eye alive - Oospora pustulans present.

Eye alive - Oospora pustulans absent.

Eye dead - Oospora pustulans present.

Eye dead - Oospora pustulans absent.

An eye was considered to be alive if any of the buds showed signs of growth. Oospora pustulans was considered present if conidiophores were visible on the buds or around the base of the buds. In the case of dead eyes with Oospora pustulans present, it could not be conclusively proved that this organism is the cause of bud death, but in the absence of any other fungal pathogen it was assumed that Oospora pustulans was probably responsible.

3. Depth of penetration of the tissue by Oospora pustulans

The first requirement for this examination is to locate sites of infection. This is simple enough when visible skin spot symptoms are evident but when this is not so, as in the earlier stages of infection, it can only be done by detecting the fungus itself. To do this the tubers were first thoroughly washed in running water and incubated for 5 days in damp cardboard boxes at 18°C. Regions of the complete tubers were then examined under a dissecting microscope for aerial conidiophores of Oospora pustulans and the located sites marked. A block of tissue round the infection site was then excised and

from this, using aseptic techniques, a longitudinal hand section of approximately 15 x 10 mm. in area and 2 mm. in thickness, was made as near as possible to the point where sporulation occurred. The section was then surface sterilised in Chloros, washed in sterile water, and incubated on a slide for 5 days at 18°C under humid conditions in the same containers as were used for the microscopic eye test. The depth of penetration of the fungus, based on the limits to which the fungus sporulated, was measured using a micrometer eye piece. The fungal growth was divided into 3 categories according to the level of sporulation, dense, sparse and infrequent (Plates 1-3).

It is true that this technique will not give as accurate an estimate of fungal penetration as histological techniques, but it has the advantages of using fresh material and is simple to carry out giving results for a large number of specimens in a relatively short space of time.

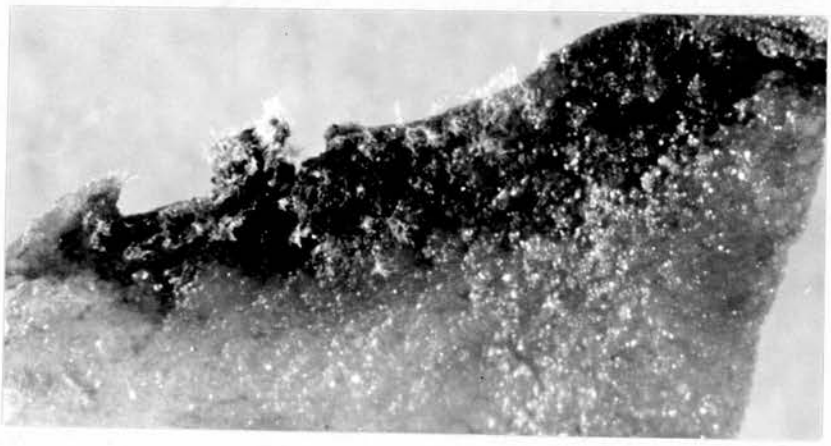
A possible criticism of the procedure may be that contamination from infection at different levels of penetration occurs in sectioning. Precautions were taken against this by sectioning from within the tuber tissue towards the surface, thus any possible contamination could only come from the lower levels and would not interfere with the depth of penetration estimation. Besides this, the aseptic techniques of flaming the razor before sectioning and surface sterilising the section before incubation would indicate that there would be little chance of the fungus surviving on the surface of the section.

Plates 1 - 3. Standards of the levels of sporulation of *Oospora pustulans* in tuber tissue.

1. Dense

2. Sparse

3. Infrequent



The assumption can be made, therefore, that the sporulation which occurs at a particular level must come from fungal material within the tissue of the section at that level. It could be argued that conidiophores which sporulate early in incubation may cause secondary infection which might in turn sporulate before examination of the section. This was tested by excising a piece of tissue from within the tuber, where there was no possibility of Oospora pustulans infection, and incubating this in close contact with a heavily sporulating piece of tissue and it was found that after 5 days no secondary infection was evident, although it did occur by about 12 days.

The technique has one important deficiency in that it is difficult to tell whether a section has been taken through the deepest part of the cup-shaped infection site. In non-visible symptom sites this is virtually impossible. In order to get an accurate comparison of the depth of penetration of the fungus in treatments it is therefore necessary to take a series of sections from different infection sites and obtain an average value of the penetration for each treatment.

4. Rate of Emergence

Weekly emergence counts were made on plots from the time the first plots emerged until the number counted each week became constant. An index of the rate of emergence of the plants from each treatment was calculated by multiplying the number of newly emerged plants counted each week by the number of days since planting and dividing the sum of these products

by the final number of plants emerged.

5. Sugar Analysis

For the investigation of the carbohydrate nutritional requirements of Oospora pustulans the fungus was incubated on media which contained the carbohydrate at the concentration of 30 g. per litre of the basal salt solution of Czapeks acid mineral solution as modified by Dox (1910) and Thom and Church (1926), quoted by Smith (1938). Analysis of total and reducing sugar content of the media from which the fungus had been filtered off was carried out using the Hanes' Ferricyanide Micromethod (Hanes, 1929).

The sugar content of tubers was determined using the same analytical technique on an aqueous extract of the sugars. This extract was taken from a 20 g. sample of the potato tissue taken from a series of cores of tissue made with a cork borer from the rose to heel end of the tubers. A fine pulp was then made of the sample with 50 ml. rectified spirit (neutral). This was slowly heated to boiling point and boiled for 5 minutes. After cooling, the solution was filtered off, the residue washed with 20 ml. aliquots of rectified spirit and the final volume of the filtrate made up to about 120 ml. This alcoholic extract of the sugars was then evaporated almost to dryness, the residue washed in a flask and made up to about 100 ml. with distilled water. This was then heated to boiling point to precipitate proteins which were then filtered off and the filtrate made up to about 150 ml. with distilled water,

this constituting the aqueous extract of the sugars in the tuber sample.

EXPERIMENTAL WORK

SECTION A

Factors at lifting and in storage in relation to skin spot development, with particular reference to the effects of level of soil moisture and temperature and humidity conditions in storage.

Introduction

While effective control of skin spot can be obtained by seed tuber disinfection with an organo-mercurial compound at lifting, such a measure is not easy to apply in practice and there is, therefore, a need for further means of reducing this problem. The successful development of possible cultural methods of control will depend upon a fuller knowledge of the factors underlying disease development. Among the factors which determine the level of skin spot development may be included, from previous work, the level of inoculum of the fungus associated with the growing crop, moisture content of the soil, the temperature and the stage of crop maturity at the time of lifting and the temperature and humidity conditions of storage. High disease incidence has been associated with above average rainfall at lifting time and lower than average temperatures in the first three months of storage (Boyd and Lennard, 1962), while reduced levels of infection have been obtained by early haulm destruction or early lifting of a crop

(Boyd, 1957). Greeves and Muskett (1939), however, found that time of lifting had no effect and although Allen (1957) suggested that tubers nearing maturity are most susceptible to skin spot, Nagdy (1962) found little difference in susceptibility to skin infection between immature and mature tubers.

In view of the limited or conflicting nature of the results of much of this previous work a series of experiments was carried out to investigate further soil conditions, time of haulm destruction and lifting and storage conditions in relation to skin spot development. In addition an examination was made of the infection progress in storage in relation to humidity and temperature conditions.

Experimental Work

A.1. The effects of factors at lifting and in storage on skin spot development.

1. Introduction

This experiment was designed to investigate the time of haulm destruction, time of lifting and storage treatment on skin spot development, the effect of boxing treatments at varying periods after harvest and clamp storage on skin spot development and the progress of fungal infection of tubers stored under clamp conditions.

2. Materials and Methods

Field scale trials were carried out at Highfield Farm, East Lothian, in two consecutive seasons, 1965-66 and 1966-67. In both growing seasons crops of the variety King Edward, grown for seed purposes, were used and the respective fields had not had crops of potatoes for six years. The seed for each crop was planted, following commercial bulk storage and grading into bags, in 28 in. drills at 11 in. spacing. The results of assessments of skin spot infection on the planted seed indicated a higher level of infection on seed planted in 1965 than on that planted in the following year when there was correspondingly less blanking (Table 1).

Table 1. Levels of skin spot infection and the degree of blanking of the planted seed in growing seasons 1965 and 1966.

Growing Season	Surface Infection Index	Eye Infection Index	Percentage Blanking
1965	30.2	70.8	17.1
1966	4.7	42.5	3.1

In both growing seasons the crop was subdivided into adjacent plots for haulm destruction with sulphuric acid at three different dates, while in the second year an additional plot was left unsprayed, allowing natural senescence to take place. The haulm destruction treatments and the plot areas for the two years were as follows:

Date of haulm Destruction		Plot Area	
1965	1966	1965	1966
23 Aug.	23 Aug.	4 drills x 400 yd.	13 drills x 300 yd.
6 Sept.	6 Sept.	"	6 "
23 Sept.	20 Sept.	"	13 "
-	Natural Senescence	-	6 "

Lifting was carried out at 5 times during each year: 23rd August, 6th and 23rd September, 6th and 19th October in 1965, and 23rd August, 6th and 20th September and 4th and 17th October in 1966, and with the exception of the first date in each year, when the clamp storage treatment was omitted, tubers were subjected to three storage treatments, clamped, boxed, and boxed and disinfected. In 1965 an additional treatment, boxed

and washed, was also included. In this year each plot for a given date of haulm destruction was sampled just prior to haulm destruction and at each subsequent lifting date to provide tubers for the various storage treatments, whereas in the second year sampling was carried out just prior to haulm destruction, two weeks later and at the final lifting date. Tubers from boxed treatments were lifted by forks and those for clamp storage by elevator digger.

Tuber yield assessments were made two weeks after each date of haulm destruction by sampling 10 x 5 yd. strips of drill within the appropriate plot. At the same time the incidence of blight was assessed by examining 100 tuber samples.

Boxed tubers were placed in standard chitting trays holding about 35 lbs. using two boxes in 1965 and three boxes in 1966 for each treatment. Any obviously diseased or damaged tubers were discarded. The disinfection technique was carried out by dipping the boxes in a tank containing 10 gallons of the organo-mercury solution used, allowing them to drain and stacking them with the other boxed treatments. The tubers were not washed before dipping. The disinfectant used was a readily available proprietary organo-mercury compound, ethoxy-ethyl-mercuric-chloride (e.e.m.c.) used at the recommended rate of 1 lb. to 20 gallons of water (150 ppm Hg) for a half minute dip. Various other disinfectants were also tested at the recommended concentrations. The washing treatment was carried out by hosing the tubers in boxes, allowing the boxes to drain and then stacking them with the other boxed

treatments. Tubers in all the boxed treatments were kept in an insulated store. The clamps were made up to 10 cwt. capacity and covered with layers of straw and finally with soil. However, larger clamps, of 1 ton capacity, were made in the early October lift to provide a source of tubers for periodic removal and boxing treatments. Minimum and maximum temperatures in the box store were taken at weekly intervals and from within the covering of the clamps at monthly intervals. In early spring samples of 50 tubers were taken from each replicate of all boxed treatments and from each of the clamp treatments and assessed for level of skin spot infection.

Statistical analysis of the effect of time of lifting, time of haulm destruction and storage treatment was carried out by the comparison of certain treatments, using Student's "t" test and analyses of variance from which general conclusions could be drawn.

From the 1 ton clamps made up in early October (haulm destruction carried out two weeks before lifting) samples of tubers were removed at intervals during the storage season and subjected to the various boxing treatments, i.e. boxed, boxed and washed (1965 only) and boxed and disinfected. At the times of removal in 1966 samples were also examined for depth of penetration of the fungus. In addition to the studies at Highfield, investigations on the effects of periodic removal and boxing from clamp storage were also carried out in 1964-65 at Langhill Farm, Midlothian, where a 1 ton clamp of tubers of the variety King Edward was made up in early October 1964 and

samples of tubers removed periodically and boxed, boxed and washed or boxed and disinfected. In all cases skin spot assessments were made in March at the end of the storage period and the effects of the various treatments determined by factorial analyses of variance of the data.

Finally from each clamping treatment tubers were sampled at lifting and at the end of the storage period for microscopic eye tests of fungal infection.

The results are considered as follows:

1. The effects of time of lifting, time of haulm destruction and storage treatment on skin spot development.
2. The effects of boxing treatments at varying periods after harvest and clamp storage on skin spot development.
3. Progressive surface and eye infection and depth of penetration of eyes by Oospora pustulans during clamp storage.

3. Results

3.1. The effects of time of lifting, time of haulm destruction and storage treatment on skin spot development

3.1.1. Tuber Yield and Incidence of Blight

Tuber yield assessments were made two weeks after each date of haulm destruction and, although the data revealed a high level of variation within treatments, the results gave evidence that, in 1965, there was no increase in tuber production from 23rd August whereas, in the following year, yield increments occurred until at least the early part of

September, this relating essentially to an increase in ware yield (Table 2).

Table 2. Tuber yield and blight incidence in relation to time of haulm destruction in growing seasons 1965 and 1966.

Date of haulm destruction		Yield (tons per acre)				Blight infected seed tubers (per cent)		Haulm decay (per cent)	
		1965		1966					
1965	1966	Ware	Seed	Ware	Seed	1965	1966	1965	1966
23 Aug.	23 Aug.	1.8	8.9	2.6	11.8	7.0	18.6	5	5
6 Sept.	6 Sept.	1.0	7.4	3.4	11.6	14.8	20.6	50	25
23 Sept.	20 Sept.	1.7	8.8	3.8	12.6	23.9	22.9	100	100
-	natural maturity	-	-	4.0	12.3	-	19.7	-	100

The table also shows the incidence of tuber blight (on seed tubers) and the percentage of haulm decay at each date of haulm destruction. In 1965 tuber blight infection was only 4.5 per cent on 23rd August and increased progressively as the season advanced to 17 per cent in October, where haulm destruction took place on 23rd August and to 33 and 35 per cent for later dates of haulm destruction (Table 3). In 1966, the corresponding figure in August was 21.5 but no further increase was apparent and time of haulm destruction had no marked effect on tuber blight from final blight assessments made in October.

Percentage of blight infected tubers in relation to time of lifting following haulm destruction on 23rd August, 1965 and 1966.

Year	Lifting Date	
	23rd Aug.	17- 19 Oct.
1965	4.5	17.0
1966	21.5	16.7

Table 3. Incidence of blight on tubers, lifted in October 1965 and 1966, in relation to time of haulm destruction.

Date of haulm destruction		Percentage of blight infected seed tubers	
1965	1966	19 Oct. 1965	17 Oct. 1966
23 Aug.	23 Aug.	17.0	16.7
6 Sept.	6 Sept.	33.0	22.0
23 Sept.	20 Sept.	35.0	19.3
-	natural maturity	-	19.7

The data are of interest in showing a contrasting pattern of yield and tuber blight development in the two years. In 1965 delayed haulm destruction not only gave no yield advantage but appreciably detracted from the value of the crop in terms of tuber blight incidence. In 1966, however, delayed haulm destruction allowed increased tuber production to take place without any apparent increase in the proportion of blighted tubers, although this was high from the beginning of the experimental period.

Foliage blight development in 1965 followed primarily an extended Beaumont Period in July. Further periods in August and early September were associated with a rapid progression of haulm decay during the early part of September. At this time of active fungal development on the foliage the considerable increase in tuber infection, in late August and

early September, may be related to the occurrence of high rainfall and to the washing of a heavy spore load into the soil. In 1966 extended Beaumont Periods occurred in June, bringing about early foliage infection which was aggravated in this experiment by the proximity of a discard pile giving rise to dense growth bearing sporulating blight lesions. Heavy rainfall in June may, in turn, have brought about early tuber infection. Dry conditions in July appeared to check the further spread of disease and allowed the growth of healthy foliage so that by 23rd August haulm blight was only 5 per cent. It was notable that infection was mainly confined to the stems. An observed high frequency of heel-end infection on tubers suggested that disease had arisen from spore passage down the stems rather than from spore showers from the leaves. Heavy rainfall accompanied by Beaumont Periods in August may have allowed further foliage and tuber infection to take place but it is suggested that dry conditions in September checked further disease development and the death of foliage in early September was associated with high winds on about 6th September rather than blight infection.

The factors accounting for the apparent variation in yield development between the two years are less evident. Temperature and sunshine records for late August and early September showed no advantage in growing conditions during 1966 although rainfall was much higher over the period in 1965. It is also difficult to account for an early cessation of tuber growth in 1965 in terms of degree of development of haulm blight.

3.1.2. Skin Spot Development

From the results of the surface and eye infection assessments (Appendices I and II) it can be seen that there was little difference between the general levels of infection for the two years despite the much higher level of inoculum on the planted seed in 1965.

The main treatment effects were as follows:

(a) Time of Haulm Destruction

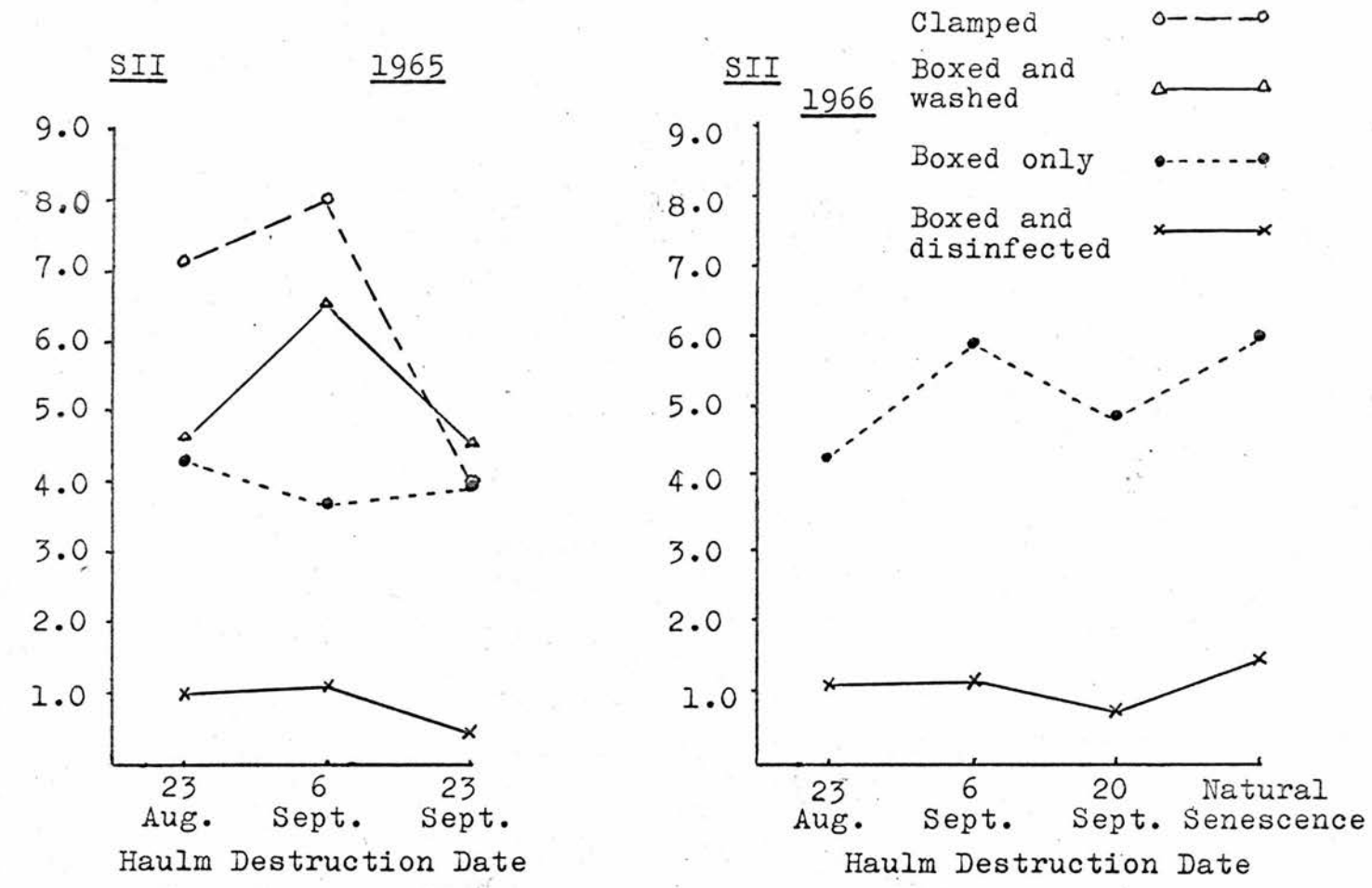
Time of haulm destruction was found to have no significant effect on the subsequent level of skin spot development in comparisons of the results for different dates of haulm destruction for tubers lifted at the same time and stored in the same way (Appendix V a-e). Thus for tubers lifted in mid-October in both years and subjected to different storage treatments, the levels of surface and eye infection showed no obvious or consistent trends in relation to time of haulm destruction (Fig. 1 and Appendix III).

(b) Time of Lifting

Since time of haulm destruction had no significant effect on skin spot development, time of lifting effects for the various storage treatments could be directly compared and the values of surface and eye infection for each time of lifting averaged for the different times of haulm destruction are represented on Figure 2 and Appendix IV.

In the case of clamp storage time of lifting had no significant effect on the level of infection which tended to

Figure 1. Skin spot surface and eye infection indices in relation to time of haulm destruction and storage treatment of tubers lifted in mid-October 1965-66.



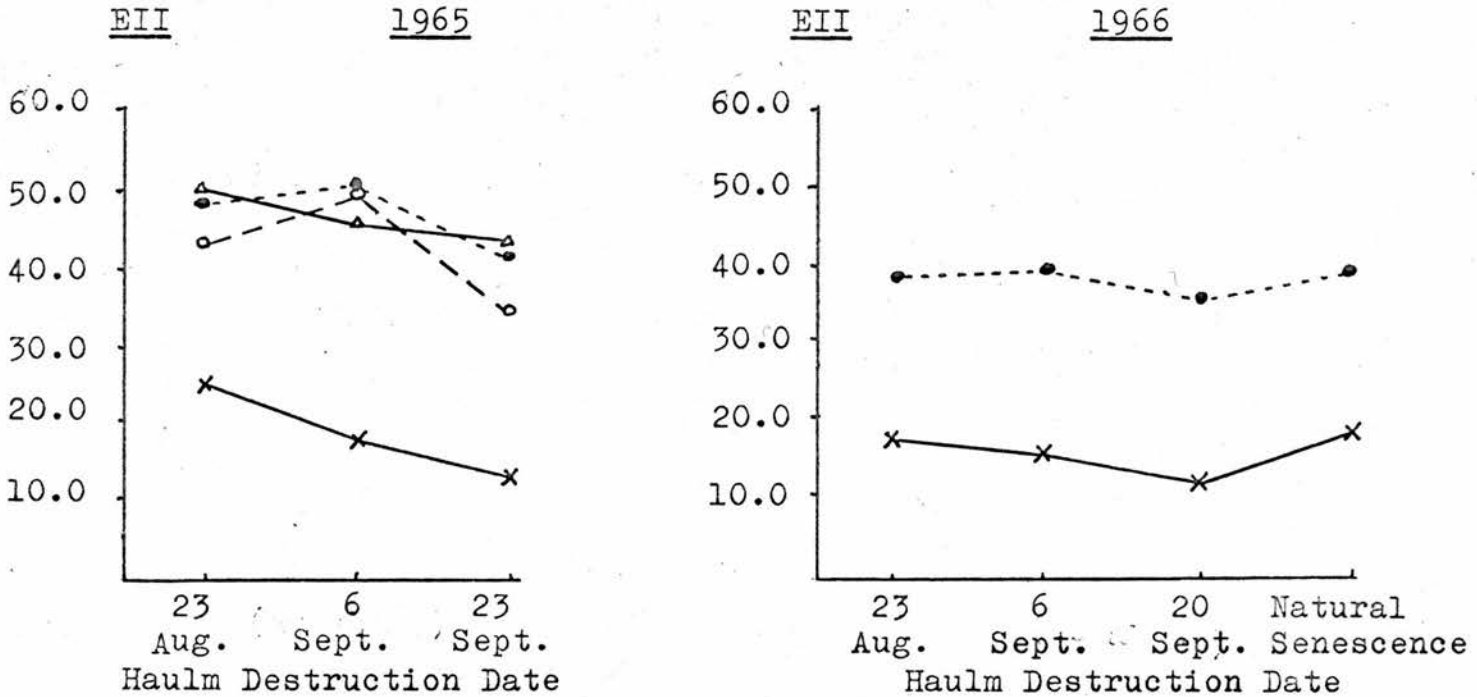
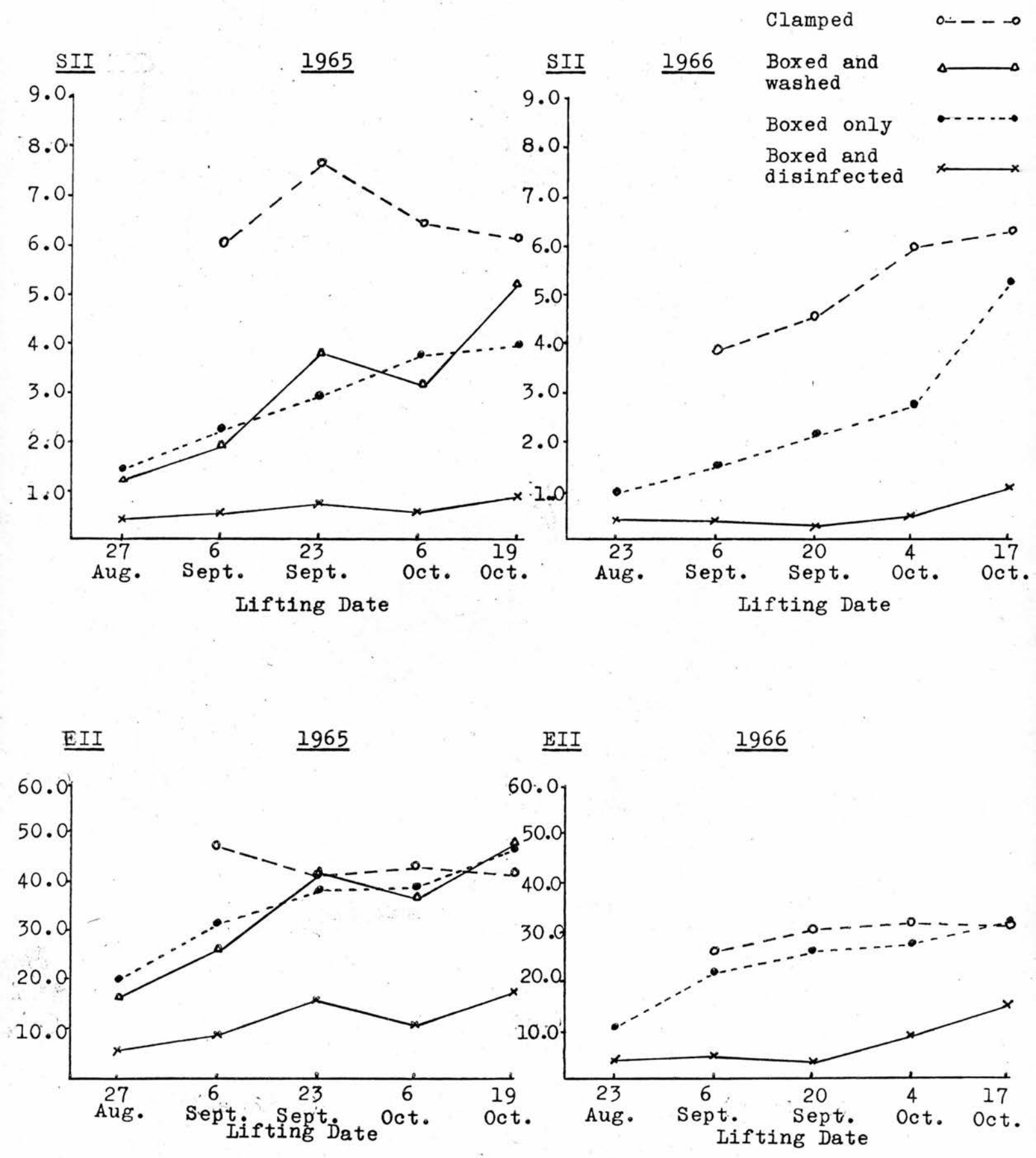


Figure 2. Skin spot surface and eye infection indices averaged for different times of haulm destruction, in relation to time of lifting and storage treatment of tubers.



be generally high (Appendices V d-i and VI). With earlier times of lifting, however, boxing significantly reduced the amount of disease which developed compared with that from clamp storage, but where lifting was delayed infection increased to a level similar to that of clamp storage (Appendix VII). Disinfection with an organo-mercury dip at lifting provided the most effective and consistent measure of control with a tendency towards better results from earlier lifting, significant in eye infection (Appendix VIII), but non-significant in surface infection (Appendix V b, h, i).

In comparing the results between years, while the levels of infection were generally similar the infection levels for the September and early October lifting times were higher in 1965 than in 1966. Previous workers have shown that among the factors encouraging skin spot development are above average rainfall at lifting time and lower than average temperatures in the first few weeks of storage. In the two years in question store temperatures and clamp temperatures were about the same until mid-November (Appendix XIV), but rainfall in September was twice the monthly average in 1965, but only half the average in 1966. This appeared to offer an explanation for the higher infection levels for these lifting times in 1965.

(c) Clamping

Where tubers were clamped, early lifting failed to give any significant reduction in subsequent skin spot development, although the 1966 figures showed a non-significant trend to

this effect. The results of boxing and boxing and disinfecting suggested a progressive establishment of infection in the field as the season advanced. This development did not seem to be markedly checked when tubers were lifted and stored in clamps, where humidity conditions presumably remained similar to those in the field. This high humidity with clamp storage, along with low winter temperatures, will always tend to favour infection, as is established.

(d) Boxing

The level of disease control obtained by boxing may be attributed to the exposure of tubers, after lifting, to dry conditions which would check fungal activity. However, there was a significant increase in the level of infection in boxed tubers as lifting time was delayed and only in early lifted tubers was there any reasonable reduction in skin spot infection compared with that in clamp storage. The results showed boxing to be as effective as boxing and disinfecting in reducing infection when tubers were lifted on 23rd August. After this lifting date, however, the efficiency of boxing compared to boxing and disinfecting in reducing infection became progressively lower (Appendix V f - n). Possible explanations for this effect are that higher ambient temperatures in storage after earlier lifting times rendered boxing more effective or that in later lifted tubers the disease establishment in the field had already reached a stage where it was no longer checked by changing humidity. With respect

to the boxing treatment Figure 2 shows that eye infection was not reduced to the same extent as surface infection by earlier lifting. It may be that eye infection was further advanced earlier in the season than surface infection because of the delicate nature of the eye tissues and the absence of a cork periderm around the eye. The fungal activity in the eye tissues, therefore, may have been less susceptible to the humidity changes brought about by boxing. It may also be that, due to the complex structure of eyes, pockets of high humidity might have remained for some time after boxing while the surface had dried. A third possible explanation is that the eye infection index may have been less sensitive than the surface infection index in detecting differences between clamped tubers and boxed tubers. As discussed in the General Materials and Methods section, there is a tendency for this index to underestimate moderately infected tubers and over-estimate slightly infected tubers.

(e) Boxing and Washing

This treatment was carried out only in 1965 and was seen to follow a similar pattern to boxing alone. There appeared to be no advantage in washing compared to boxing alone and in the subsequent year the treatment was discarded.

(f) Boxing and Disinfecting

Boxing and disinfecting tubers with the e.e.m.c. fungicide at lifting appeared the most efficient means of controlling skin spot. There was a trend, significant only with eye

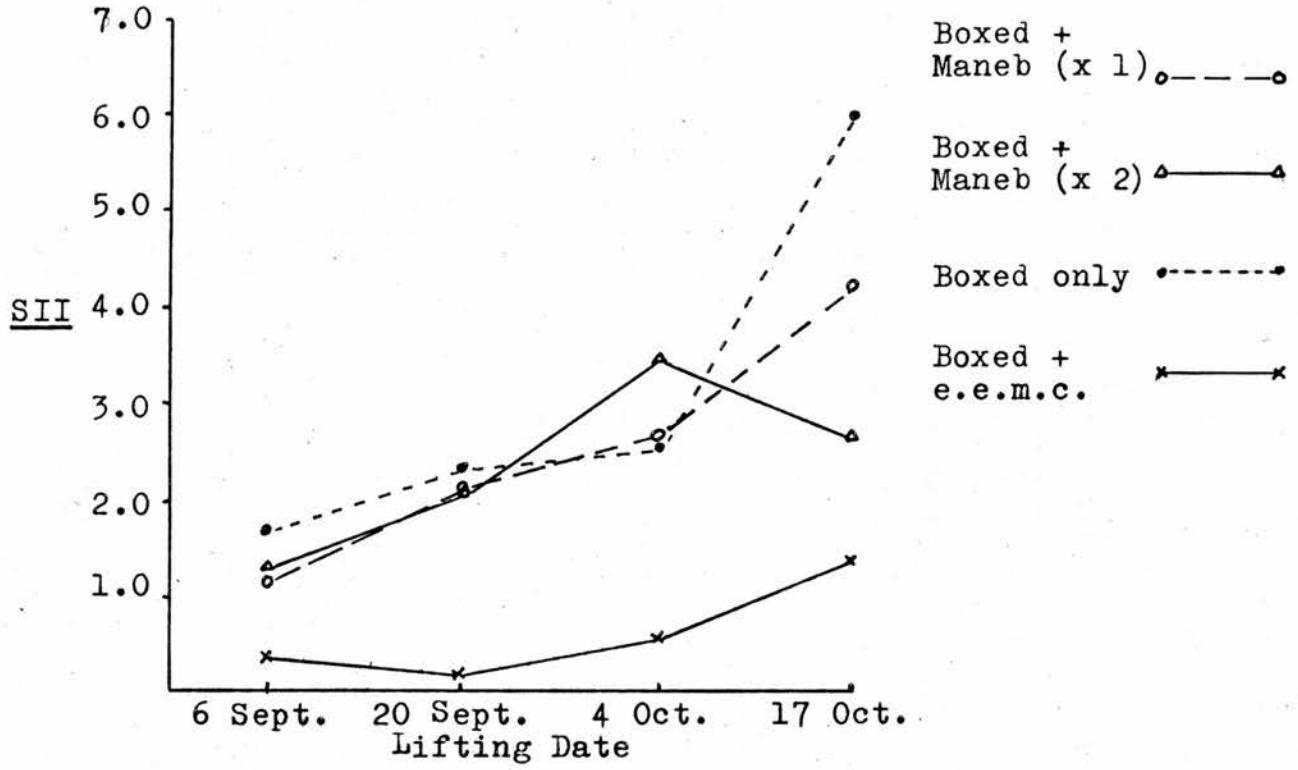
infection, towards more effective control with earlier lifting which might have reflected progressive fungal infection as the season advanced. However, the level of control of skin spot, even from the latest lifting time, was as good as that which boxing achieved with the earliest lifting time.

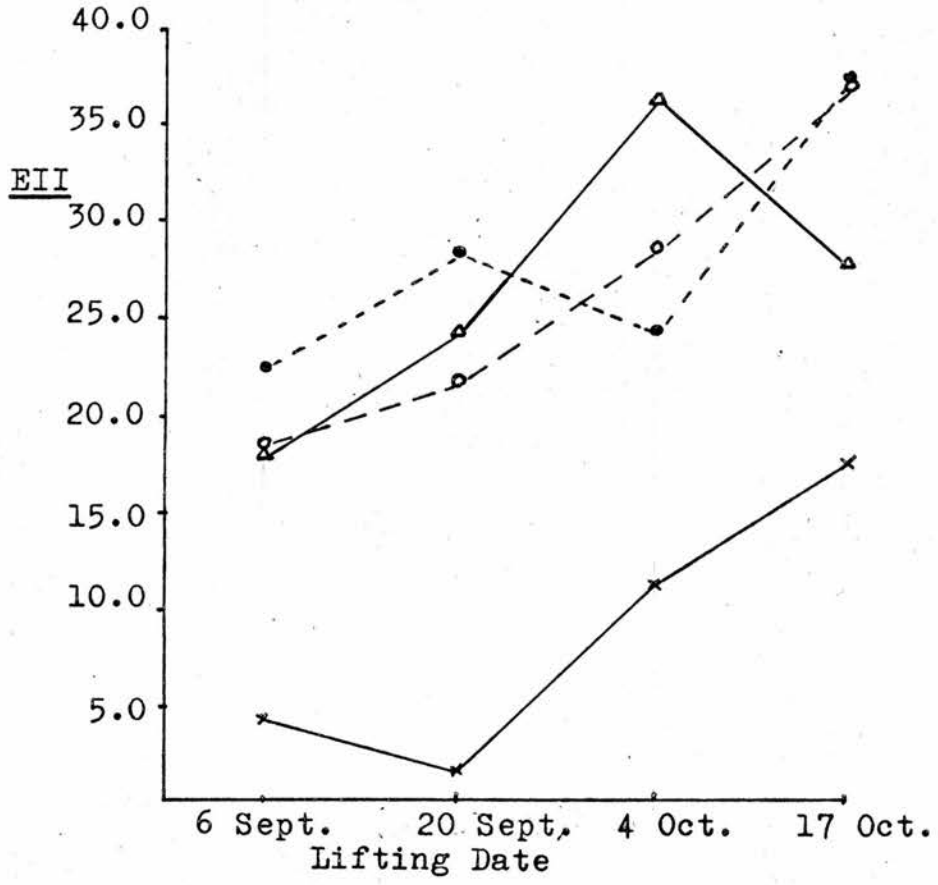
Where fungicides other than an organo-mercury solution were used for disinfection the results (Table 4) showed that the e.e.m.c. compound reduced skin spot to a greater extent than any of the other compounds investigated. Maneb in 1965 gave some reduction compared with boxing alone, but in 1966 this was only evident when the solution was at double the recommended concentration with the addition of a wetter. The only other compound to exert any kind of control was 3% copper sulphate but this caused extensive chemical damage on the tubers. In 1966, Maneb was investigated in more detail by using two different concentrations and dipping at each lifting time. The effects of Maneb compared with e.e.m.c. and boxing alone (Fig. 3 and Appendix IX) showed that e.e.m.c. was consistently more efficient than the other treatments in controlling skin spot while there was little difference between the two Maneb strengths which proved no better than boxing in reducing skin spot. The improvement in the efficiency of Maneb by the application of a wetter might warrant further investigation, but the results for the other disinfecting treatments suggest no possibilities for their use as alternatives to the organo-mercury solution.

Table 4. Effects of various fungicidal treatments of tubers lifted on 6th October 1965 and 17th October 1966, on the subsequent skin spot development.

Fungicidal Treatment	Skin spot development			
	1965		1966	
	S.I.I.	E.I.I.	S.I.I.	E.I.I.
e.e.m.c. ½ lb in 10 gal ½ min.	0.7	13.6	1.4	17.4
Maneb (80 per cent DP) (at ¾ lb in 10 gal) 3 min.	1.7	25.6	4.2	36.8
Maneb (80 per cent DP) (at 1½ lb in 10 gal 3 min. and wetter)	-	-	2.6	27.9
Captan (50 per cent WP) ¾ lb in 10 gal 3 min.	3.4	51.5	-	-
Thiram (80 per cent DP) ¾ lb in 10 gal 3 min.	3.8	44.4	-	-
3% Copper sulphate 3 lb in 10 gal 3 min.	-	-	2.6	24.4
Fentin acetate 1 lb in 10 gal 3 min.	-	-	4.1	31.4
Copper oxychloride 2½ lb in 10 gal 3 min.	-	-	4.4	38.9
Blitane 1½ lb in 10 gal 3 min.	-	-	4.9	32.2
Boxed alone	3.8	40.5	6.0	37.5

Figure 3. Comparison of the effects on skin spot development of disinfected with e.e.m.c., with Maneb (at 2 concentrations) and with boxing only at different times of lifting - 1966.





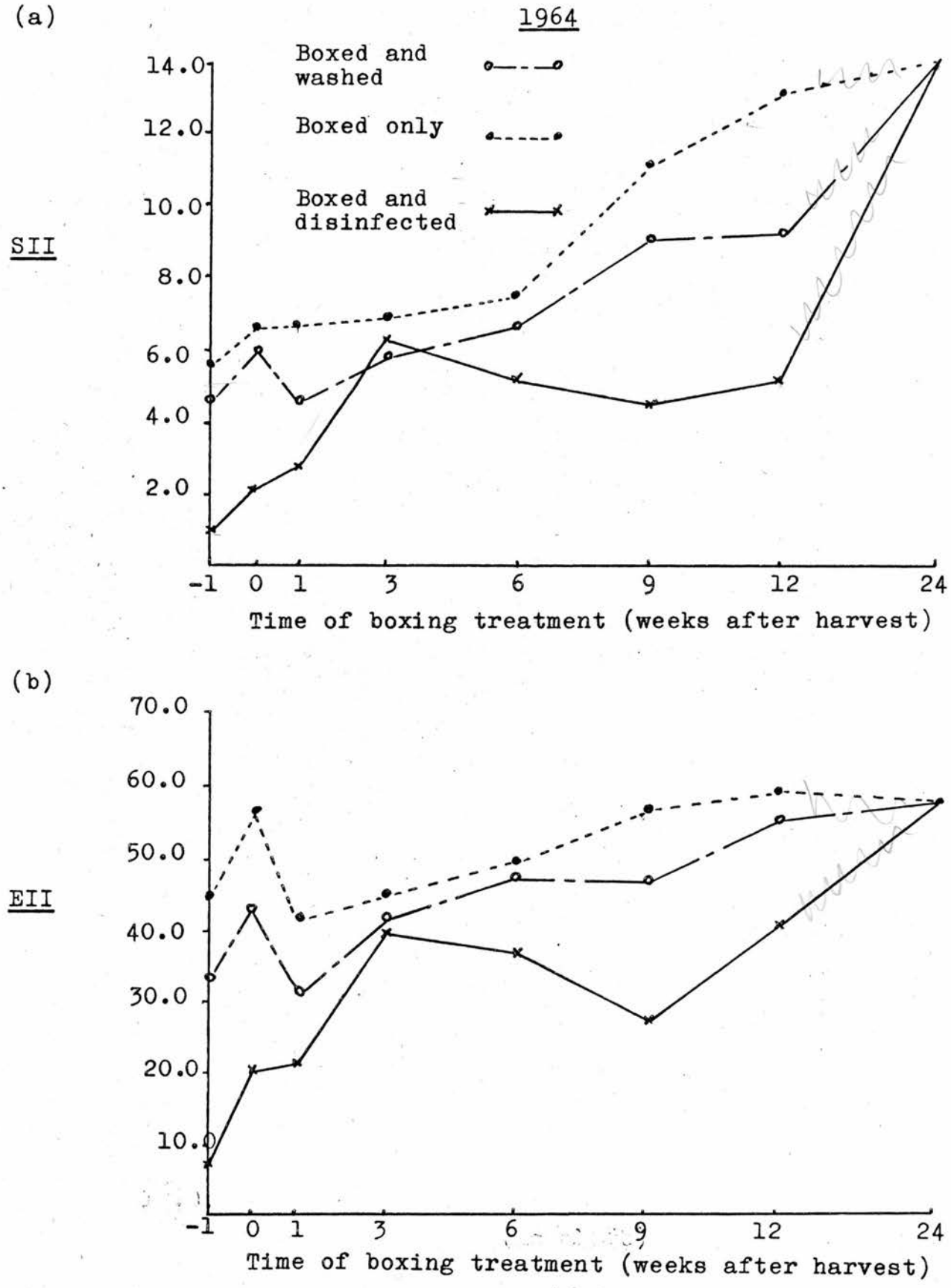
3.2. The effects of boxing treatments at varying periods after harvest and clamp storage on skin spot development

The surface and eye infection indices in relation to the various boxing treatments carried out at different times and after clamp storage are shown on Figures 4a-f and Appendices X and XI. In general, the results indicated that boxing and disinfecting provided a greater degree of control of skin spot than either boxing alone or boxing and washing for all times of treatment, its effectiveness decreasing with delay in removal from the clamp (Appendices XII and XIII). From Figures 4c-f it may be seen that in seasons 1965 and 1966 a reasonable level of control, with respect to surface and eye infection was maintained up to six weeks after lifting, but in 1964 (Figs. 4a and b) boxing and disinfection was only effective until three weeks after lifting compared with boxing or clamp storage.

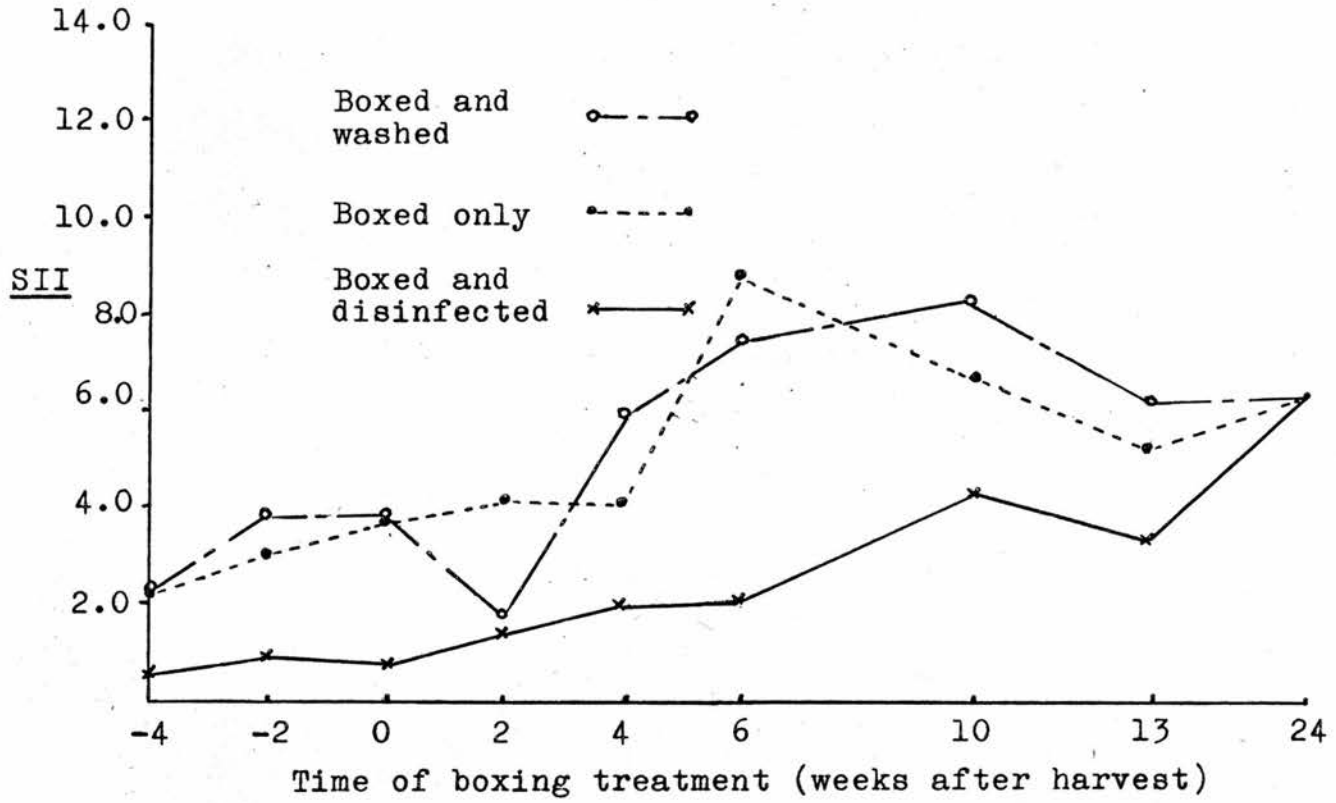
In comparison with clamp storage, boxing at lifting gave a slight reduction in surface infection level in two of the three years (1965, 1966) and boxing after up to six weeks in clamp storage appeared to afford some control in one of the three years (1964). In general, however, boxing at the normal time of lifting or following clamp storage failed to provide an effective or consistent means of control of surface infection and gave no reduction in level of eye infection compared with that of continuous clamp storage.

There seemed to be little difference between the effects

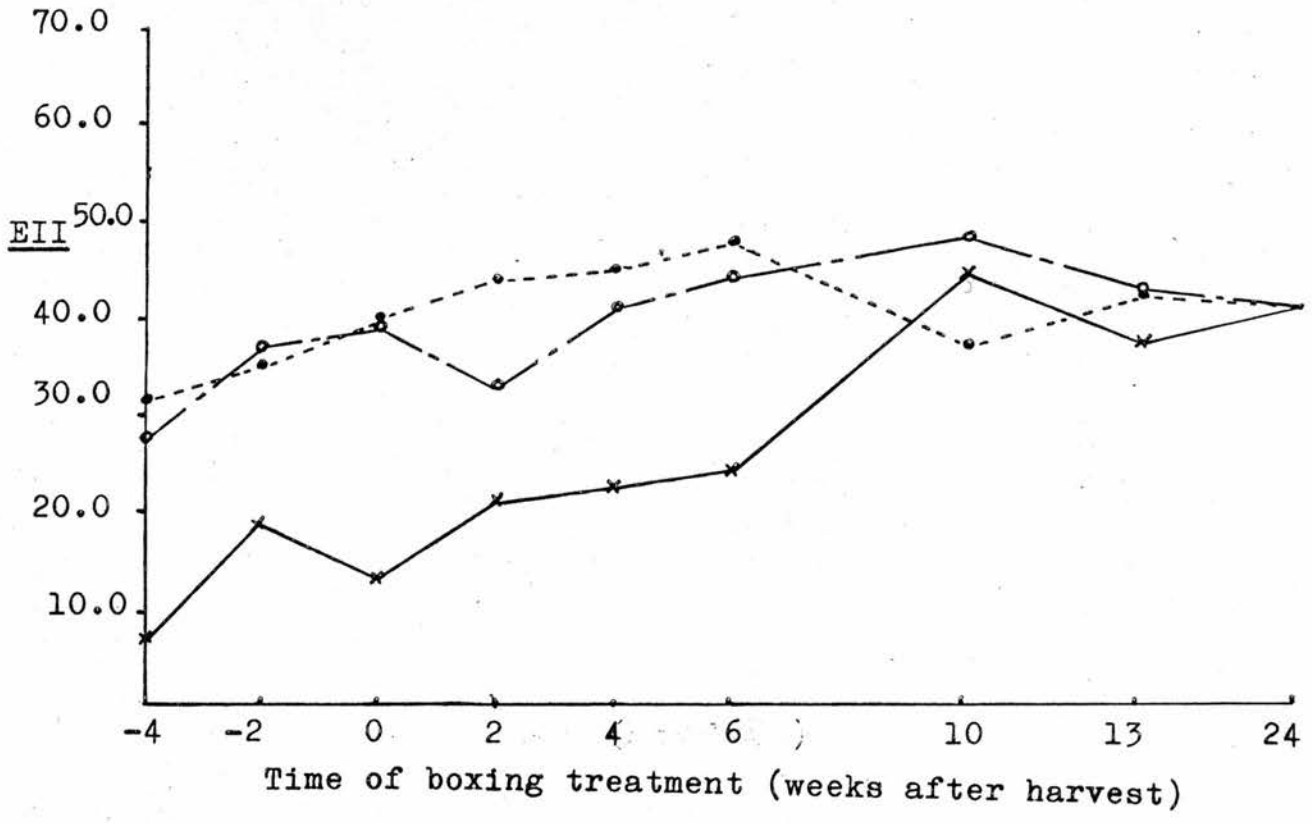
Figure 4. Skin spot surface and eye infection indices in relation to different box storage treatments following clamp storage from lifting in early October 1964-1966.



(c) 1965


14.0
12.0
10.0
SII
8.0
6.0
4.0
2.0
Boxed and washed
Boxed only
Boxed and disinfected
-4
-2
0
2
4
6
10
13
24
Time of boxing treatment (weeks after harvest)

(d)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EII 50.0
40.0
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-4
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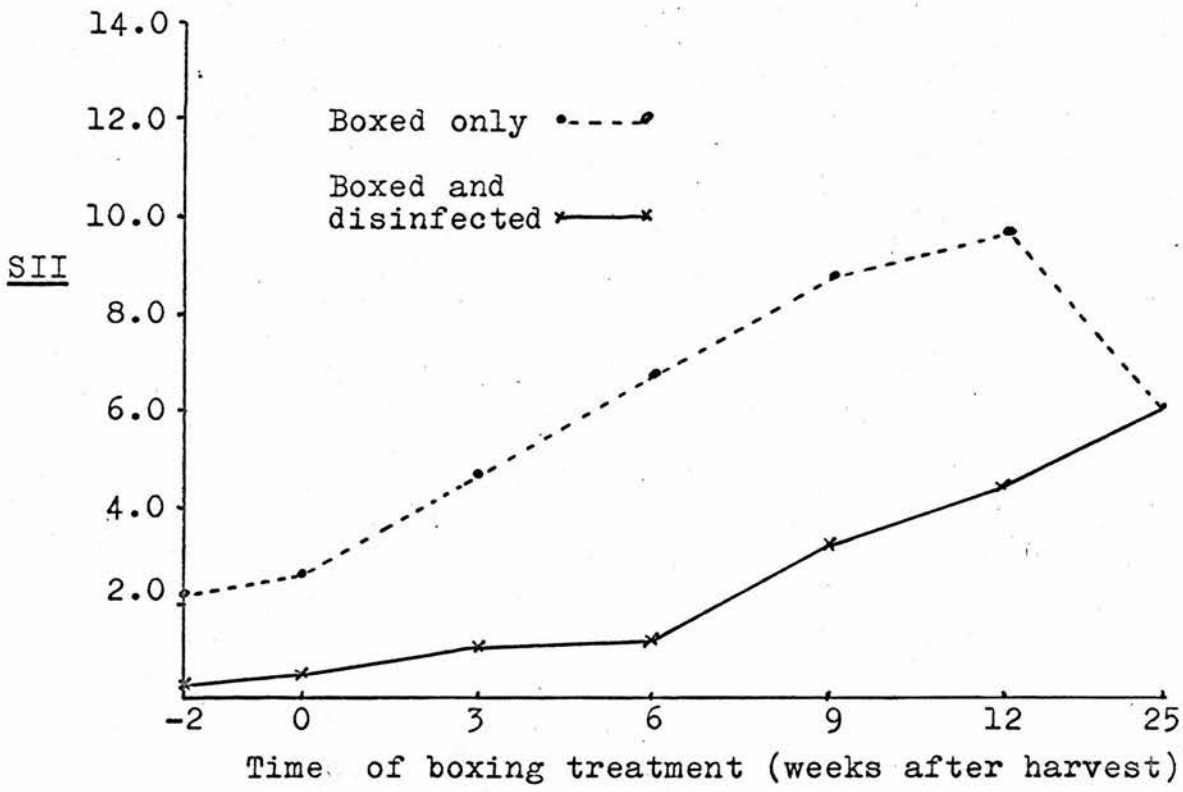
24
Time of boxing treatment (weeks after harvest)

(e)

1966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Boxed only
Boxed and disinfected
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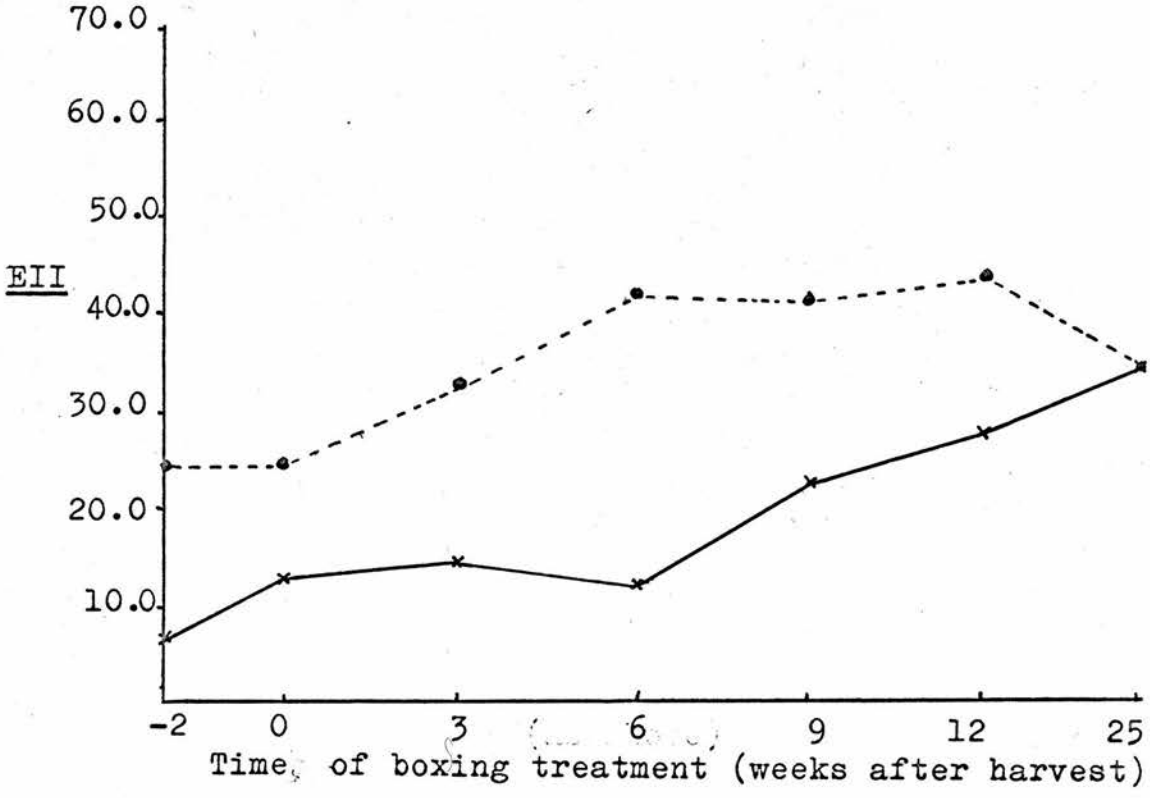
Time of boxing treatment (weeks after harvest)

(f)


70.0
60.0
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40.0
30.0
20.0
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EII
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9
12
25
Time of boxing treatment (weeks after harvest)

of boxing alone and boxing and washing.

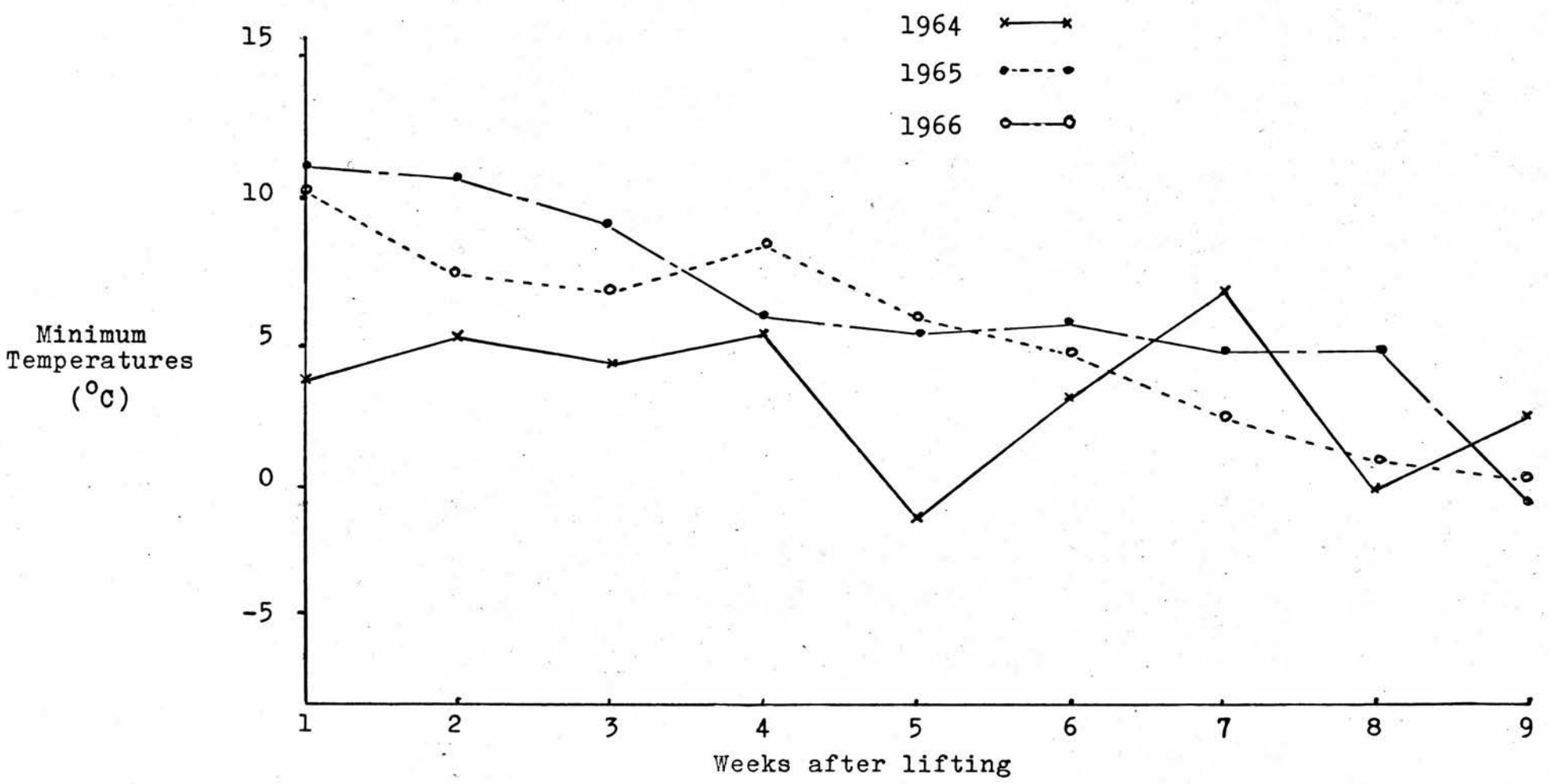
In comparing skin spot development in the three seasons, in 1964 there was a higher level of infection than in 1965 and 1966. Factors which are associated with high levels of skin spot infection of a crop are a heavy infection on the planted seed, above average rainfall over the lifting period and below average temperatures in the first three months of storage. Table 5 shows the infection levels of the planted seed and the rainfall figures for September in each year.

Table 5. Level of skin spot infection of planted seed and the total rainfall in September in growing seasons 1964, 1965 and 1966.

Growing season	Skin spot infection		Total rainfall in September
	SII	EII	
1964	7.4	41.0	2.77 ins.
1965	30.2	70.8	4.55 ins.
1966	4.7	42.5	1.47 ins.

These figures show that a high level of seed inoculum and heavy rainfall in September would favour skin spot infection in 1965, while the 1964 and 1966 figures were relatively low. However, temperature records (Fig. 5 and Appendix XIV) showed that in the first six weeks of storage the weekly minimum temperatures were considerably lower in 1964 than in 1965 and 1966 although subsequent to this the temperature levels were similar for the rest of the storage period. The high

Figure 5. Weekly minimum temperatures during the first 9 weeks of storage, after lifting, in an insulated shed 1964-1966.



infection level in 1964 may, therefore, be related to the low temperatures in the first six weeks of storage. Although no temperature figures are available it can be assumed, since the clamp was in the same area as the storage shed, that in 1964 the temperature figures were correspondingly low in the clamp as in the shed and under these conditions the fungal infection appeared to have become so established that worthwhile chemical control by removing tubers from the clamp and disinfecting could only be achieved within three weeks after harvest compared with six weeks in 1965 and 1966. These results suggested that temperature levels in the few weeks immediately after normal lifting time were critical in terms of skin spot development and the effectiveness of measures taken to reduce infection.

3.3. Progressive surface eye infection and depth of penetration of eyes by Oospora pustulans during clamp storage.

The results of microscopic eye tests carried out at lifting and at the end of clamp storage for the various times of haulm destruction and times of lifting treatments (Tables 6 and 7) indicated generally increase in the number of dead eyes and in the number of eyes infected with Oospora pustulans by the end of the storage period. No differences were evident between the various treatments.

The average depth of penetration of the pathogen and the average depths of the different levels of sporulation at

various periods after clamp storage from lifting are shown on Figure 6 and Appendix XV with Plates 4 and 5 showing the penetration at the 6 and 20 week stages after lifting. The infection of tubers removed from clamp storage at the same periods and subjected to boxing or boxing and disinfecting are recorded on Figures 7a and b and Appendix XVI. These results showed that skin spot development could be kept reasonably low by disinfecting with organo-mercury up to six weeks after harvest and this may be associated with a relatively low depth of penetration of the fungus. The failure of disinfection to provide effective control after six weeks of clamp storage may be related to the depth of penetration of the fungus having reached an advanced stage subsequent to this period. Moreover it may be seen that by six weeks infection was already established in the surface layers and this may account for the failure of boxing to give satisfactory control at this or earlier times of treatment, the fungus having already established itself to an extent that it was no longer susceptible to changing humidity conditions.

Appendix XV & XVI. Why are ex-clamp figures lower than those for boxed only?

Table 6. Eye infection, by Oospora pustulans, of tubers subjected to different haulm destruction times and lifting times, measured microscopically, at lifting time and in March after clamp storage, season 1965-66.

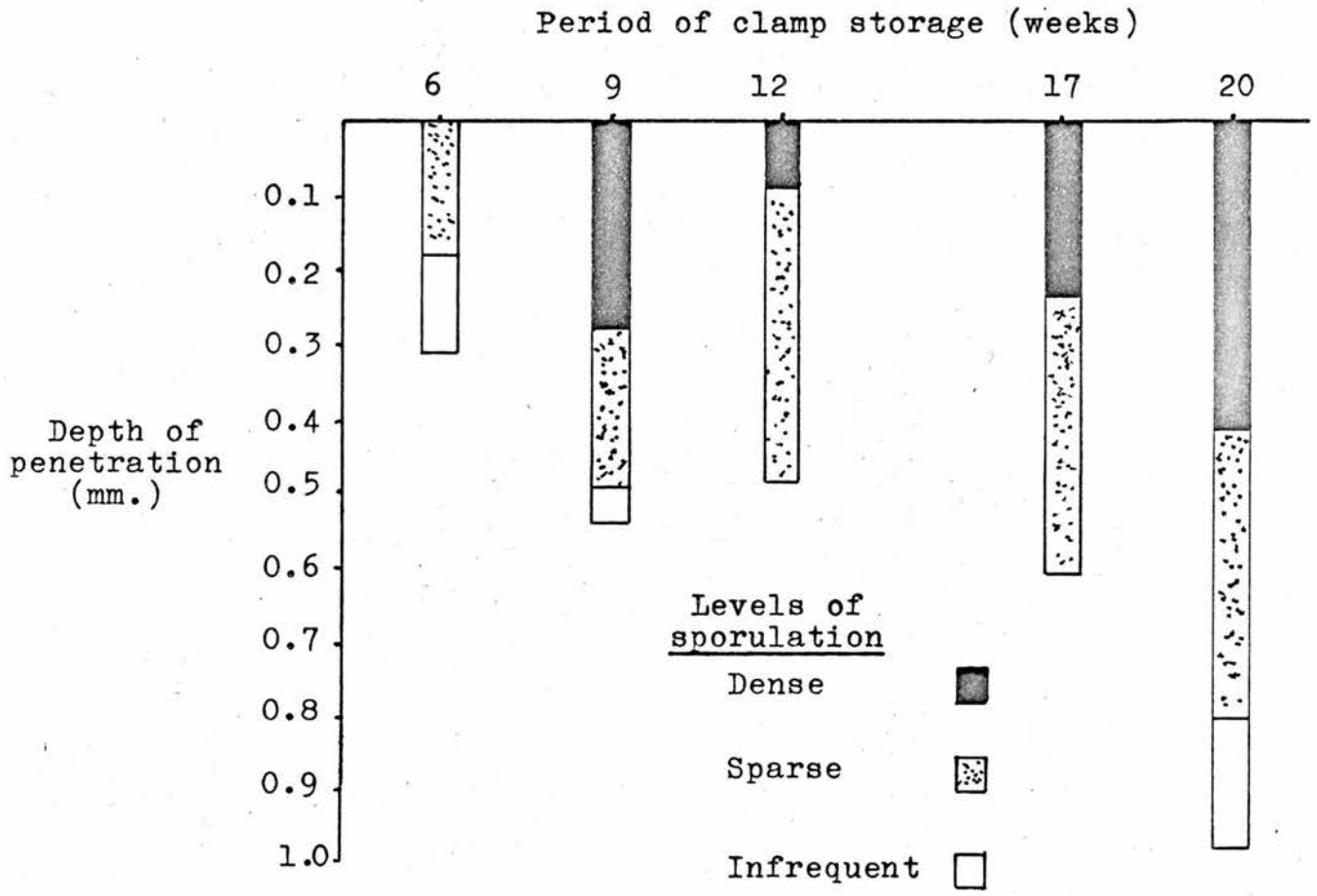
Date of Haulm Destruction	Date of lifting	Eyes dead (per cent)		Eyes showing Oospora pustulans (per cent)	
		At lifting	After clamp storage	At lifting	After clamp storage
23 Aug.	6 Sept.	16	34	18	58
	23 Sept.	24	34	38	55
	6 Oct.	15	31	23	69
	19 Oct.	44	25	19	42
6 Sept.	23 Sept.	26	24	31	44
	6 Oct.	21	32	16	68
	19 Oct.	15	34	26	62
23 Sept.	6 Oct.	18	38	24	64
	19 Oct.	14	28	23	55

Table 7. Eye infection, by Oospora pustulans, of tubers subjected to different haulm destruction times and lifting times, measured microscopically, at lifting time and in March after clamp storage, season 1966-67.

Date of Haulm Destruction	Date of lifting	Eyes dead (per cent)		Eyes showing Oospora pustulans (per cent)	
		At lifting	After clamp storage	At lifting	After clamp storage
23 Aug.	5 Sept.	1	11	19	35
6 Sept.	20 Sept.	2	14	27	26
23 Sept.	4 Oct.	1	23	22	48
natural senescence	17 Oct.	6	20	18	53

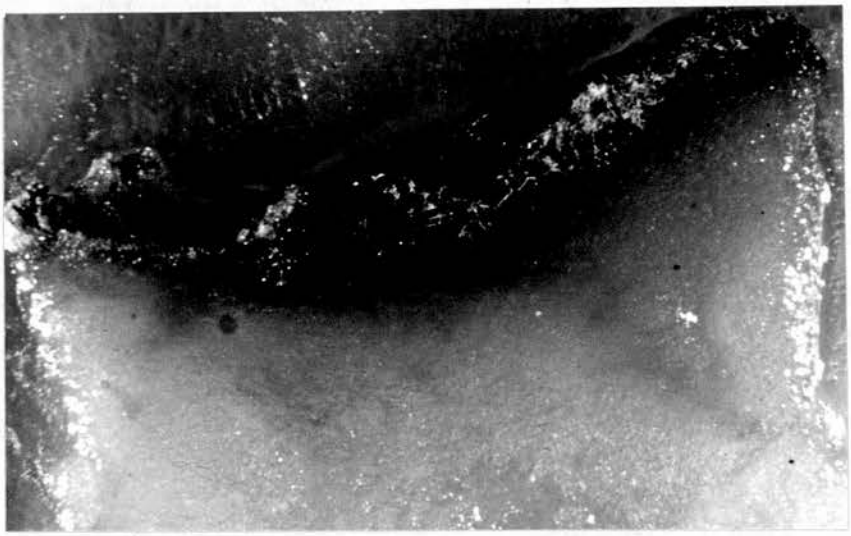
23 Sept 23% eyes dead. In App XVI 33% eyes dead
This should be explained, as % o E.I.I.

Figure 6. The average depths of the different levels of sporulation (mm.) and the total average depth of penetration of Oospora pustulans (mm.) into the tissue of tubers after various periods of clamp storage from lifting 4 Oct. 1966.



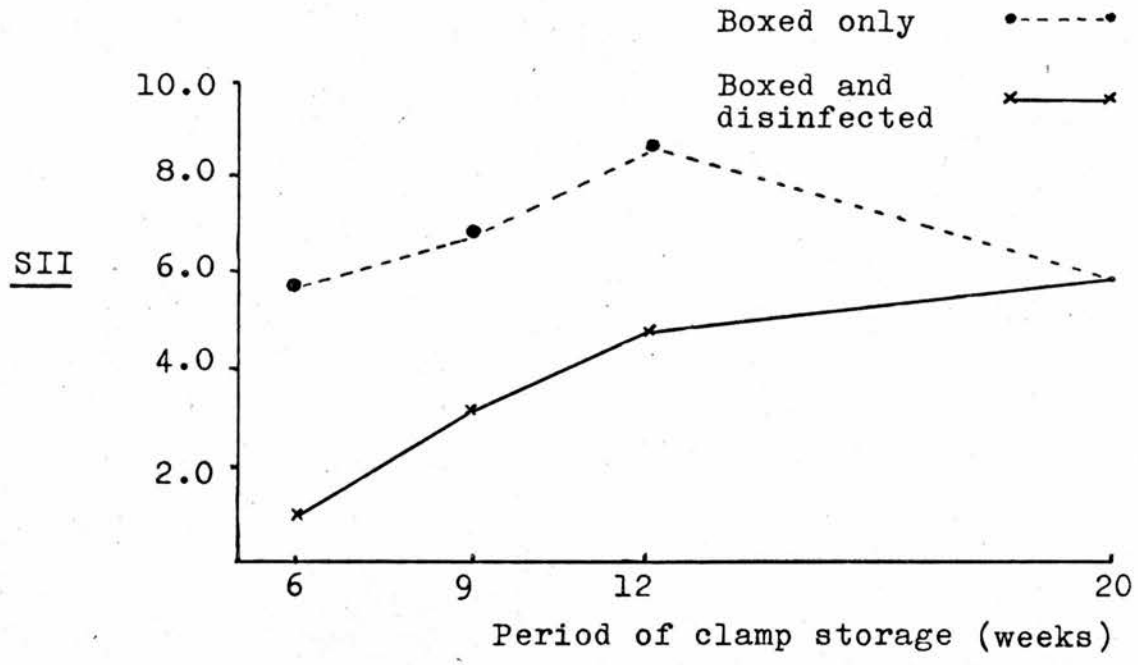
Plates 4 - 5. The depth of penetration of Oospora pustulans into tuber eye tissue at 6 and 20 week stages after lifting.

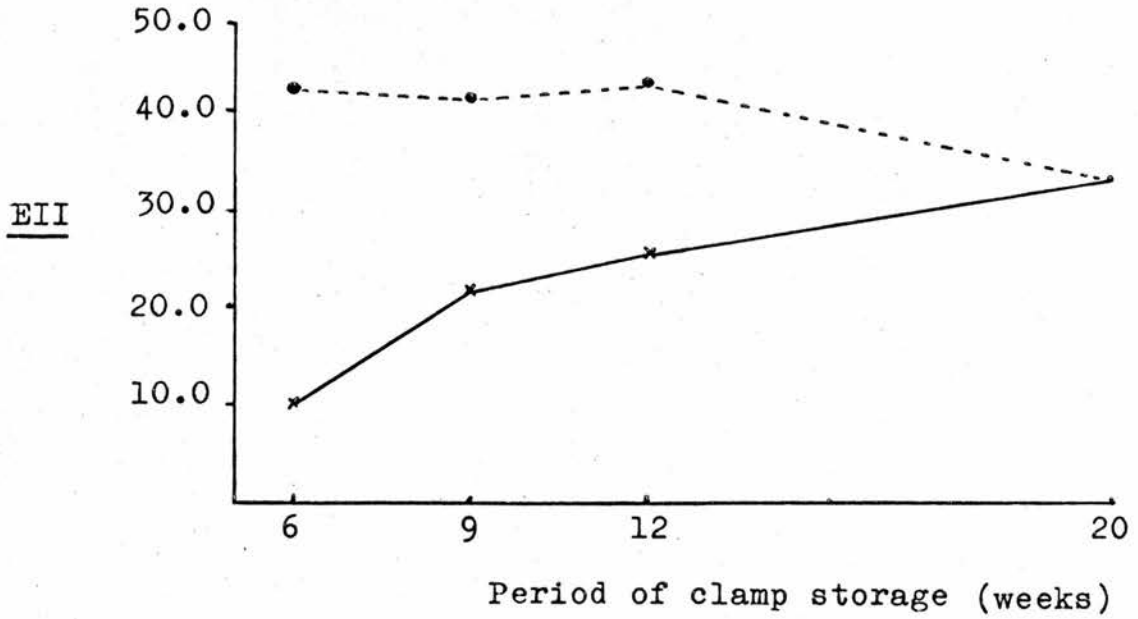
4. 6 weeks



5. 20 weeks

Figure 7. Skin spot development in tubers subjected to different box storage treatments after various periods of clamp storage from lifting 4 Oct. 1966.





A.2. The effects of varying levels of soil moisture on skin spot development.

1. Introduction

The work of Boyd and Lennard (1962) has shown that rainfall during the lifting period is an important factor in the subsequent development of skin spot. In preliminary investigations to get more precise information about a possible soil moisture effect polythene covers over the drills were used in an attempt to control soil moisture level but failed to prove effective. In this experiment an alternative technique to control soil moisture was employed.

2. Materials and Methods

Micro-plots were constructed from wooden boxes 16 in. x 10 in. area and 12 in. deep. The bottom of the boxes had drainage slots and a 1 in. layer of gravel was placed in each. The boxes were filled with a medium-loam field soil to about 6 ins. from the top and fertiliser was then applied at the rate of 100 units of nitrogen per acre. This was sprinkled on and mixed in with the soil. Two King Edward seed tubers with slight surface infection and some eyes infected were then placed in each box and covered with 6 ins. of soil. The boxes were then sunk into the earthen floor of a bird proof cage, 27 ft. x 18 ft. area and 12 ft. in height, with soil packed round each box so that normal soil temperatures could be achieved. The plots were laid out linearly in 4 blocks of

16 with a 1 ft. soil barrier between each block. Within the blocks the plots were arranged in a 4 x 4 rectangle.

The plots received normal rainfall until 4th July 1966 when the cage was covered with heavy duty polythene to prevent rain access and from then until 5th September wet treatments were applied to 2 blocks and dry treatments to the other 2. Tensiometers were placed in random plots within the blocks with the control level from the wet treatments at about 5 cm. of Hg soil moisture tension, equivalent to a soil moisture content in a medium-loam soil of about 25 per cent (Searle, 1954) and for the dry treatments at about 55 cm. of Hg, equivalent to a soil moisture content of about 5 per cent. To keep the plots at approximately these levels the wet treatments were watered at the rate of 0.91 ins. of water per plot per week and the dry treatments at 0.18 ins. of water per plot per week. In the 8 week period from 5th September until 1st November different watering treatments were applied at weekly intervals (Table 8). With the hitherto wet plots, in the first week all the plots received 0.5 ins. of water except one row of 4 plots. Treatments in the subsequent weeks were similar, only water was withheld from a further row of 4 plots each week giving a series of 8 treatments with the period without water supply ranging from 1 to 8 weeks before 1st November. With the hitherto dry plots, in the first week one row of 4 plots was watered at the rate of 0.5 ins. of water and the remaining plots kept dry and in the subsequent weeks a further row of 4 plots each week was included for watering,

Table 8. Plot watering treatments in the investigation of the effects of soil moisture on skin spot development.

Date of application of treatment	Treatments			
	Plots Wet until 5 Sept.		Plots Dry until 5 Sept.	
	Number of rows unwatered	Number of rows receiving 0.5 ins. water per week	Number of rows unwatered	Number of rows receiving 0.5 ins. water per week
5th Sept.	1	7	7	1
12th Sept.	2	6	6	2
19th Sept.	3	5	5	3
26th Sept.	4	4	4	4
3rd Oct.	5	3	3	5
10th Oct.	6	2	2	6
17th Oct.	7	1	1	7
23rd Oct.	8	0	0	8

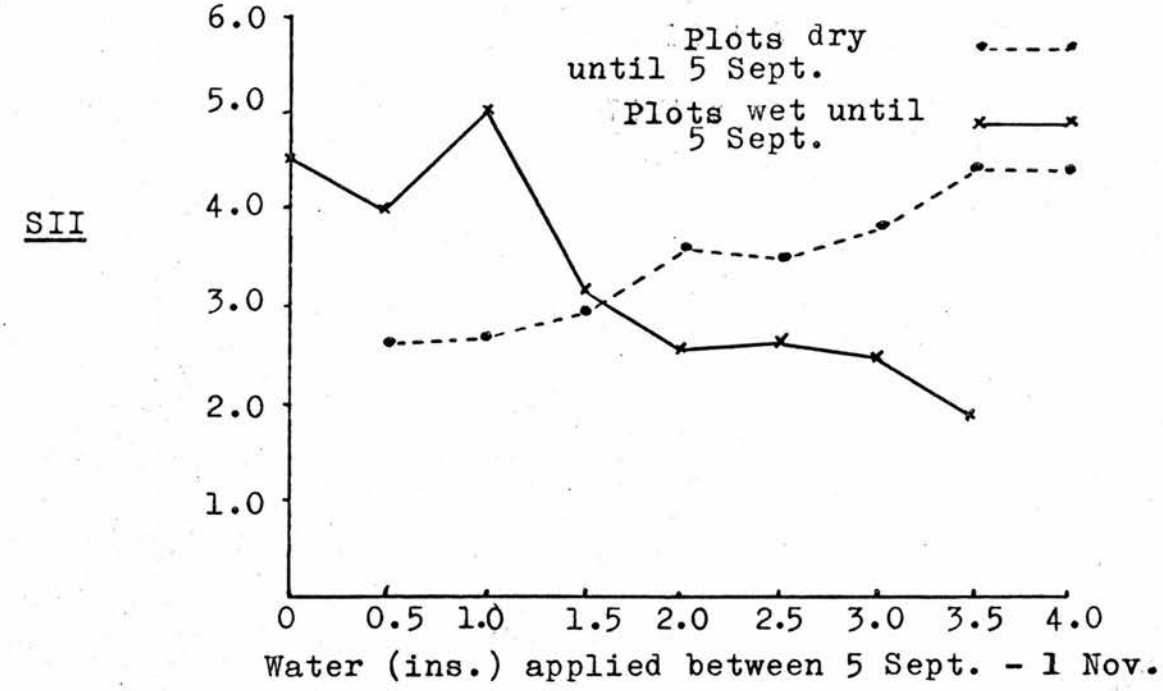
giving another series of 8 treatments with the period of no water supply ranging at weekly intervals from 1 to 8 weeks before 1st November. On 1st November the tubers were harvested from the plots, placed in net bags and stored in a clamp until March 1967 when the treatments were assessed for skin spot development. Statistical analysis of the results was carried out by applying Student's 't' test on comparable treatments.

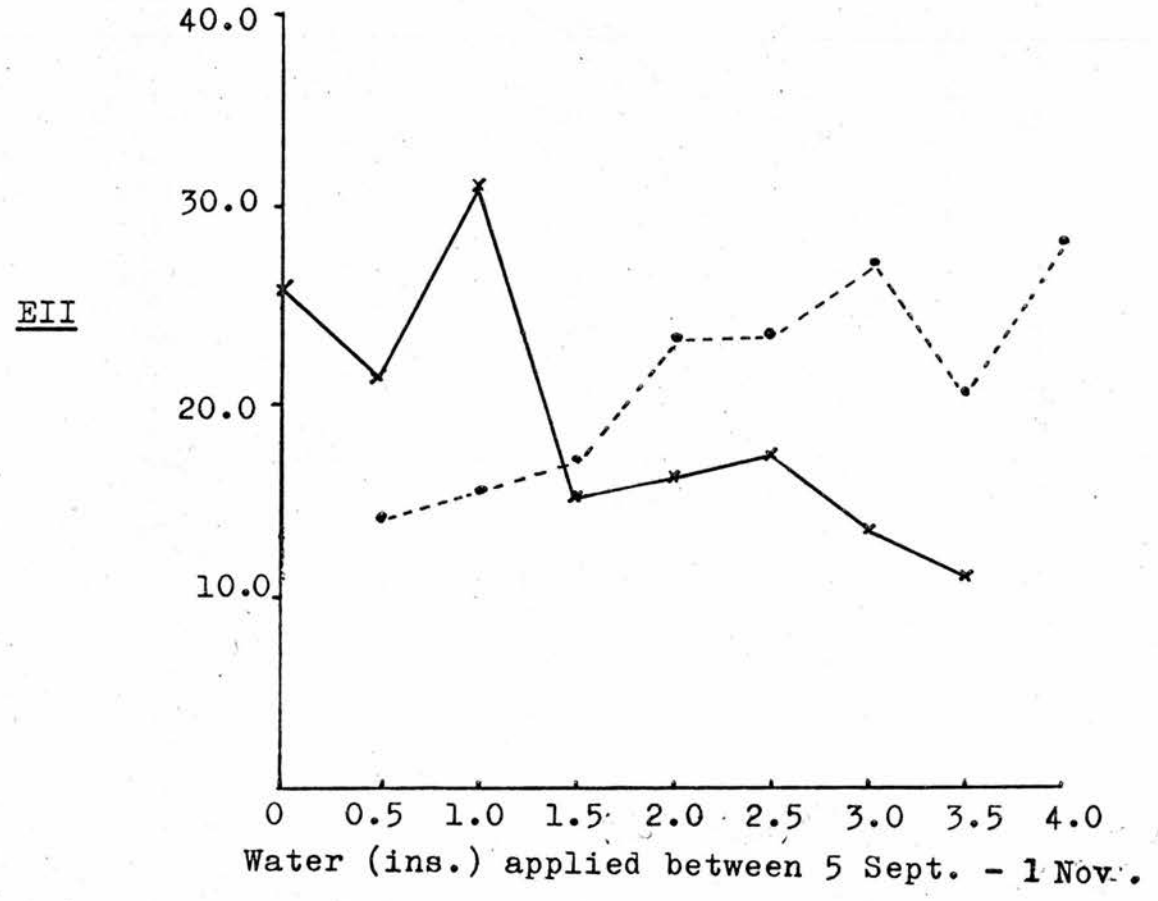
3. Results

The surface and eye infection indices for the treatments are shown on Figures 8a and b and Appendix XVII. From the plots which were dry until 5th September an increase in disease level was found as more water was applied to the soil over the 8 week period to 1st November. Although the differences were not significant (Appendix XVIII), the pattern shown was in agreement with the suggestion that higher rainfall over the lifting period is associated with higher skin spot infection. With the plots which were kept reasonably wet until 5th September, the highest level of disease developed from the treatment which received 1 in. of water in the 8 week period until 1st November. Where progressively more water was applied the disease incidence correspondingly decreased and there was a significantly lower level of disease development in the treatments receiving 2 in. of water and more compared with that where 1 in. of water was applied (Appendix XVIII).

Making a direct comparison between the water applied to the plots and normal rainfall figures over the same period it

Figure 8. Skin spot development related to different soil watering regimes - 1966.





is difficult to explain the results in the wet plot treatments since the total water applied before 5th September was more or less the same as that which an average rainfall would give and that applied to the most heavily watered treatment was about half of what an average rainfall would give over the 8 week period - 5th September to 1st November. Under these circumstances an increase in infection as opposed to an inhibition of infection might be expected. These unexpected results may have been due to the watering system and possible lack of drainage from the plots resulting in the water levels as applied in this experiment bearing little relation to similar rainfall levels in the field and those levels which showed an inhibition of disease development may have been equivalent to abnormally high rainfall in effect. Moreover the fact that the treatments were stored together in the same clamp may have masked certain real soil moisture effects since the humidity in the clamp was likely to become uniform throughout the treatments and the full effect on infection of soil moisture in the field may be related to the humidity thus produced in storage.

This applies to all other experiments where differences are found.

A.3. The effects of different storage conditions and temperature on skin spot development.

1. Introduction

This experiment was carried out to compare the effects of continuous high and low storage temperature, of storage in an insulated shed and of storage in a clamp on skin spot development.

2. Materials and Methods

King Edward seed tubers were lifted on 15th October 1964 and subjected to the following four treatments:

(a) Storage at 15°C (60°F)

(b) Storage at 4.5°C (40°F)

(c) Storage in an insulated shed (2°C - 14°C) (29°F - 53°F)

(d) Storage in a potato clamp.

For treatment (d) the tubers were held in 16 net bags each containing 50 tubers and made up into a small clamp while in the remaining treatments the tubers were stored in small cardboard boxes kept moist by spraying with water with 16 boxes of 10 tubers each per treatment. In March 1965 the tubers were examined for development of skin spot.

3. Results

There was a significant decrease (Appendix XIX) in both surface and eye infection in the tubers held at 15°C compared with that of the tubers held under the other storage conditions

which all gave high levels of infection (Table 9).

Table 9. Skin spot development in tubers under different storage conditions and temperature regimes.

Storage treatment	Skin Spot Development	
	S.I.I.	E.I.I.
Continuous at approx. 15°C	1.0	6.8
Continuous at approx. 5°C	16.1	68.1
Fluctuating storage temperature (2°C - 14°C)	11.3	66.8
Fluctuating clamp temperature (no temperature figures available)	12.6	71.2

There was, however, a high degree of sprouting in the tubers held at 15°C. The tubers held continuously at 5°C had a higher surface infection index than those held in the insulated shed and in the clamp. This may be attributed to the higher temperatures in the latter two environments in the first few weeks of storage, the temperature records of the area showing that it was not until about six weeks after storage that the ambient temperature was continuously as low as 5°C. The same difference in infection was not reflected in the eye infection indices and it may be further evidence that eye infection is further advanced than surface infection at lifting

time.

The type of pustules produced in the treatments varied, those on tubers stored at 5°C were large and sunken, the ones on tubers stored in the shed or clamp were the normal raised type and on tubers held at 15°C they were very small.

A.4. The effect of varying temperature conditions during the later stages of storage on skin spot development.

1. Introduction

This experiment was carried out to examine the effects of temperature changes in the later stages of storage on skin spot development.

2. Materials and Methods

Seed tubers of the variety King Edward were lifted on 3rd November 1966 and subjected to the following treatments.

(a) Stored at about 9°C (48°F) throughout the storage period.

(b) Stored at about 9°C (48°F) until 2nd February then stored at about 5°C (40°F).

(c) Stored at about 5°C (40°F) throughout the storage period.

(d) Stored at about 5°C (40°F) until 2nd February then stored at about 9°C (48°F).

For each treatment cardboard boxes of 10 tubers replicated 8 times were used. These boxes were kept moist by spraying periodically with water.

In March 1967 the treatments were examined for development of skin spot.

3. Results

Table 10 shows the skin spot development in the treatments and it was found that significantly less skin spot was found on tubers stored at 9°C than on those stored at 5°C during the

Table 10. Skin spot development in tubers subjected to changing temperature conditions in the later stages of storage.

Type of pustule	Storage Treatment	Skin spot development	
		S.I.I.	E.I.I.
Normal raised type	9°C continuous	4.4	40.6
Normal raised type	9°C until 2 Feb. and then 5°C	6.8	49.3
Abnormally large and sunken	5°C continuous	9.6	56.8
Abnormally large and sunken	5°C until 2 Feb. and then 9°C	9.9	60.6

whole period of storage. Transferring tubers from storage at 5°C to storage at 9°C at the beginning of February did not change the level of skin spot development compared with continuous storage at 5°C. There was a slight but non-significant increase in the level of infection where tubers were transferred from storage at 9°C to storage at 5°C at the beginning of February compared with continuous storage at 9°C (Appendix XX).

Tubers stored continuously at 5°C developed abnormally large and sunken skin spot pustules as were found in the previous experiment. This type of symptom was also produced in the tubers which were placed in warmer storage at the beginning of February, whereas the tubers kept at 9°C until the beginning of February and then placed in colder storage had normal pustules as had those kept constantly at 9°C. It

would appear, therefore, that the size of the pustule was determined by the temperature regime in the earlier storage stages.

A.5. The spread of infection by Oospora pustulans on the surface of tubers and its depth of penetration into the tissues under different environmental conditions.

1. Introduction

During the storage season 1966-67 an experiment was designed to study the surface activity of Oospora pustulans on tubers and its depth of penetration into the tissues under controlled temperature and humidity conditions.

2. Materials and Methods

Tubers of the variety King Edward were lifted on 3rd November 1966, washed and stored at 5°C until 8th November when the 8 treatments as shown on Table 11 were applied.

Table 11. Treatments used in the investigation of the spread of Oospora pustulans on the surface of tubers and its depth of penetration into the tissues under different environmental conditions.

Treatment	Infection Treatment	Humidity Level	Temperature Level
1	natural infection	Dry	10°C
2	artificial inoculation		
3	natural infection	Damp	
4	artificial inoculation		
5	natural infection	Dry	5°C
6	artificial inoculation		
7	natural infection	Damp	
8	artificial inoculation		

For the inoculated treatments the tubers were immersed for 1 minute in a spore suspension of Oospora pustulans (750,000 spores/ml.) to which 40 g. of sterile soil and 80 g. of sterile peat was added per litre in an attempt to improve the adhesion of the inoculum. Tubers for the uninoculated treatments were similarly immersed in a sterile soil and compost suspension. Following immersion the tubers were placed on grids in metal containers (18 x 18 x 18 in.) each holding 75 tubers and covered with a loose fitting hardboard lid. For the damp treatments, the atmosphere within each container was kept damp by placing a tray of water below the grid supporting the tubers and a moist pad of cotton wool and paper towelling on a grid above the tubers. In the dry treatments the tubers were simply held on the grid at normal atmospheric humidity. The containers for the different temperature treatments were placed in cabinets at 10°C (50°F) and 5°C (40°F) respectively. The humidity in the damp treatments was maintained above 90% relative humidity and in the dry treatments at about 60% at 10°C and 80% at 5°C.

Microscopic eye tests were made on the original sample of tubers and on 15 tuber samples for each treatment taken at 4 week intervals for 5 sampling dates. The eye tests were made using the complete tuber technique described in the General Materials and Methods section. An examination for the depth of penetration by the fungus was also carried out on each sampling date.

3. Results

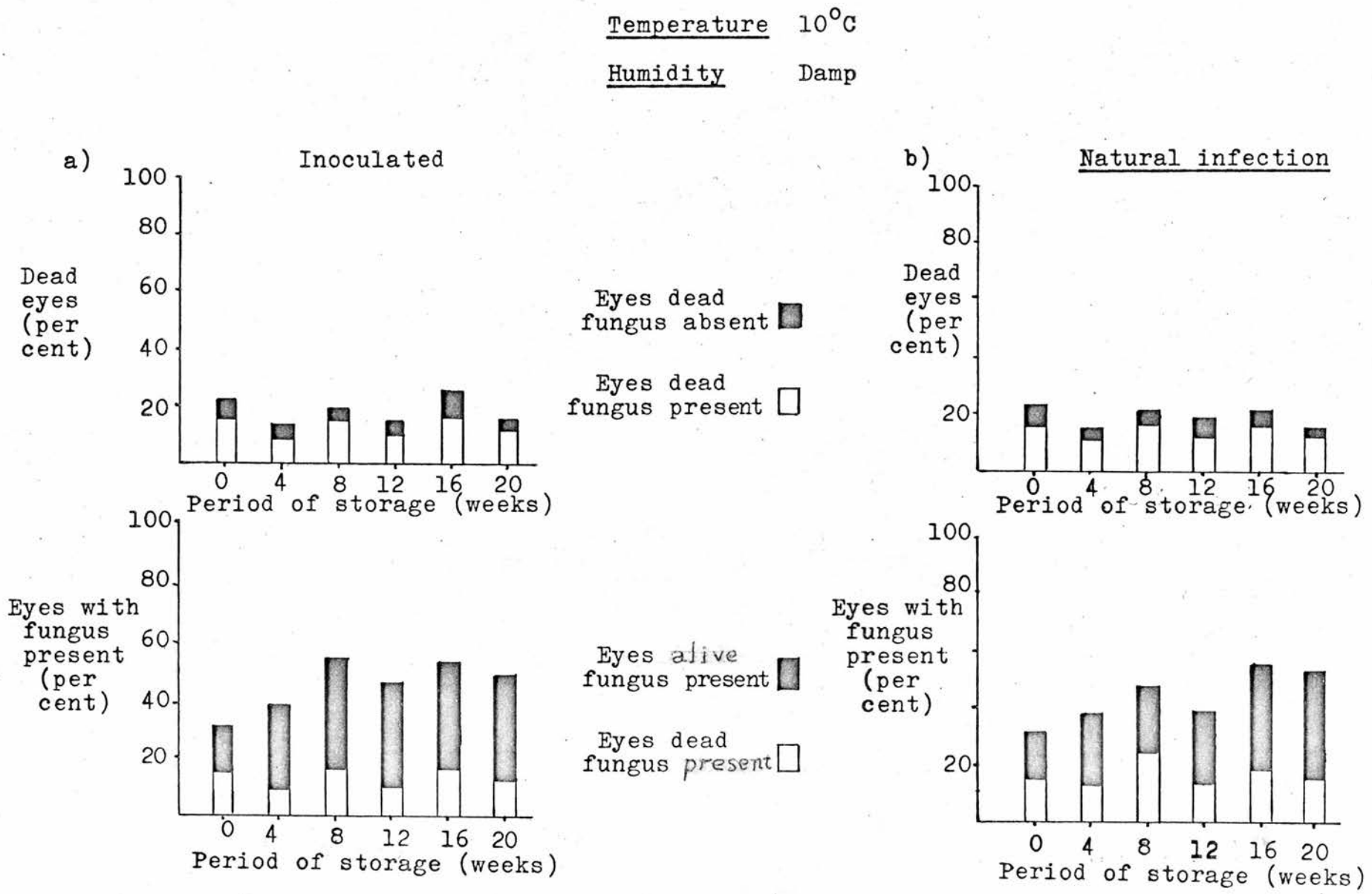
3.1. Surface Infection

The results on Figures 9a-h and Appendix XXIa, b show the levels of Oospora pustulans (on dead and live eyes) and of dead eyes (infected and non-infected by the fungus) at the different stages of storage. In all treatments there was a proportion of dead eyes with no fungal infection present which remained more or less constant.

In the 10°C treatments (Figs. 9a-d) the total number of dead eyes during the storage period showed no increase. The damp treatments at this temperature (Figs. 9a, b) gave an increase in fungal infection but the eyes thus infected were not killed. The dry artificially inoculated treatment (Fig. 9c) also showed an increase in fungal infection on live eyes, but this was very slight and considerably less than in the damp treatments. In the dry natural infection (Fig. 9d), however, virtually no increase in the fungus was apparent. The Oospora pustulans levels in the damp natural infection treatment (Fig. 9b) showed a progressive increase during storage, while in the 2 inoculated treatments (Figs. 9a, c) the level reached a peak at the 4-8 week period and did not progress from there. However, despite this early increase in infection from inoculation under these conditions there was little difference in the final levels of fungal infection for the natural infection and artificially inoculated treatments.

In the 5°C treatments (Figs. 9e-h) the level of dead eyes increased progressively during storage, was most marked

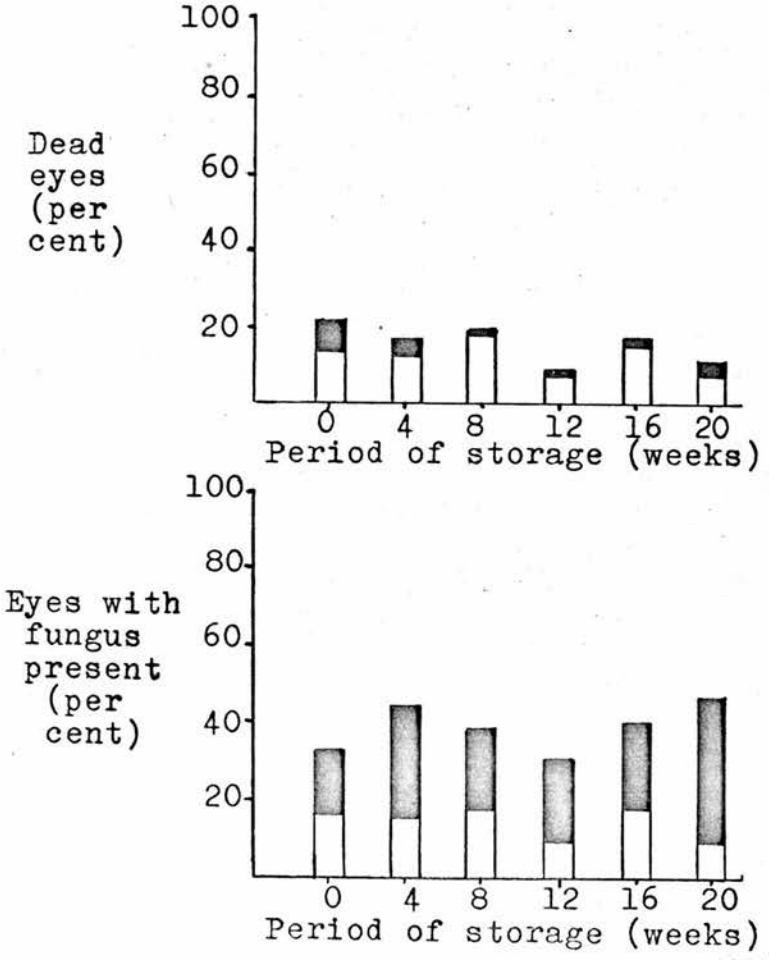
Figure 9. Microscopic eye tests, made at 4 week intervals during storage, of tubers, naturally infected and artificially inoculated with Oospora pustulans, and subjected to different humidity and temperature regimes.



Temperature 10°C

Humidity Dry

c) Inoculated



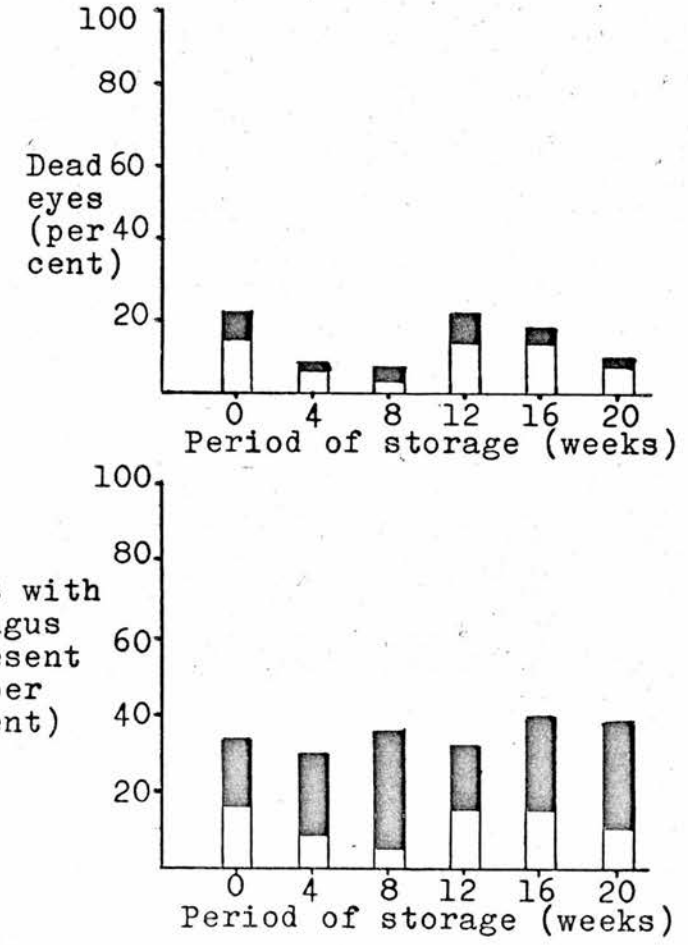
Eyes dead fungus absent

Eyes dead fungus present

Eyes alive fungus present

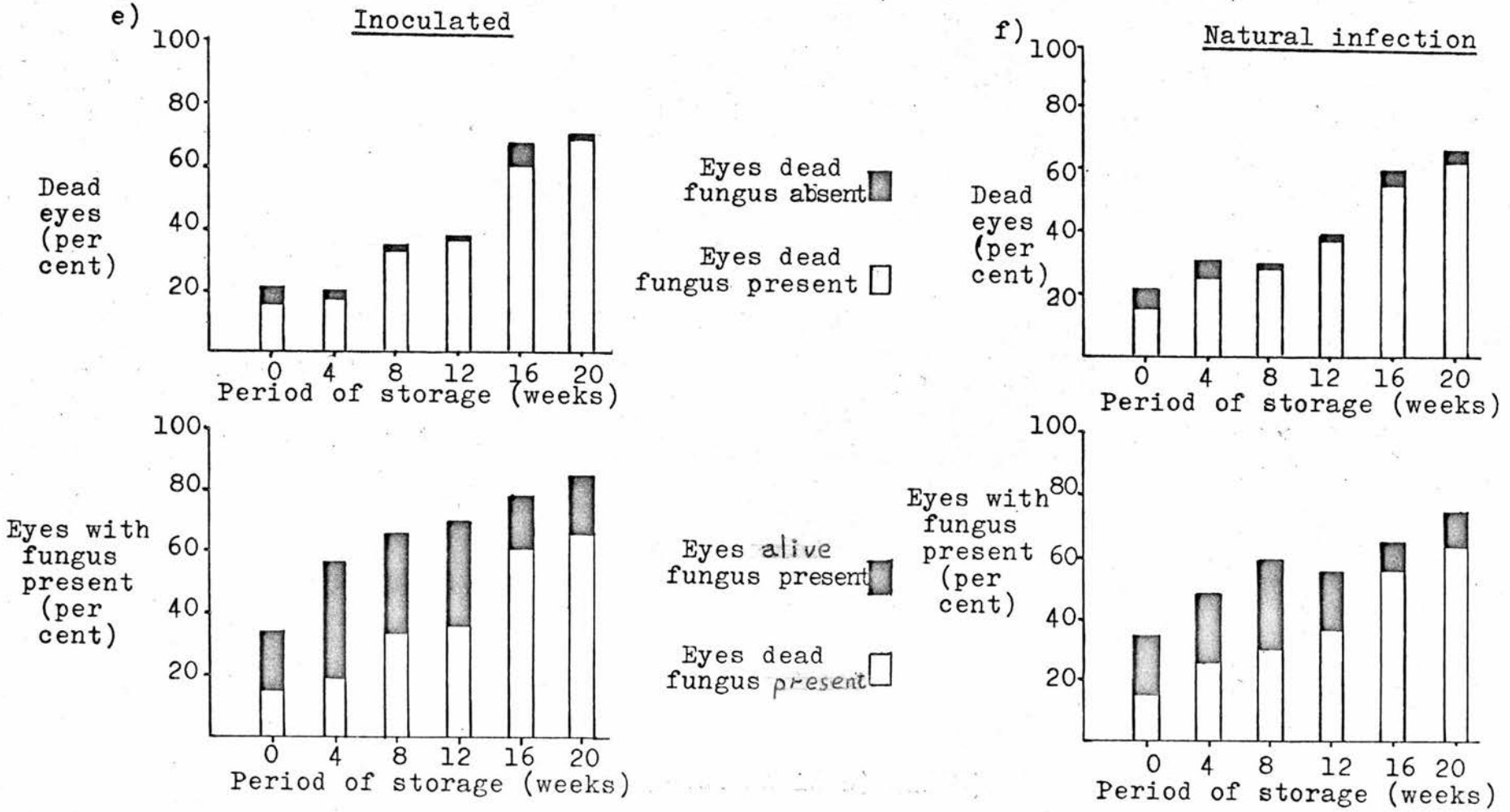
Eyes dead fungus present

d) Natural infection



Temperature 5°C

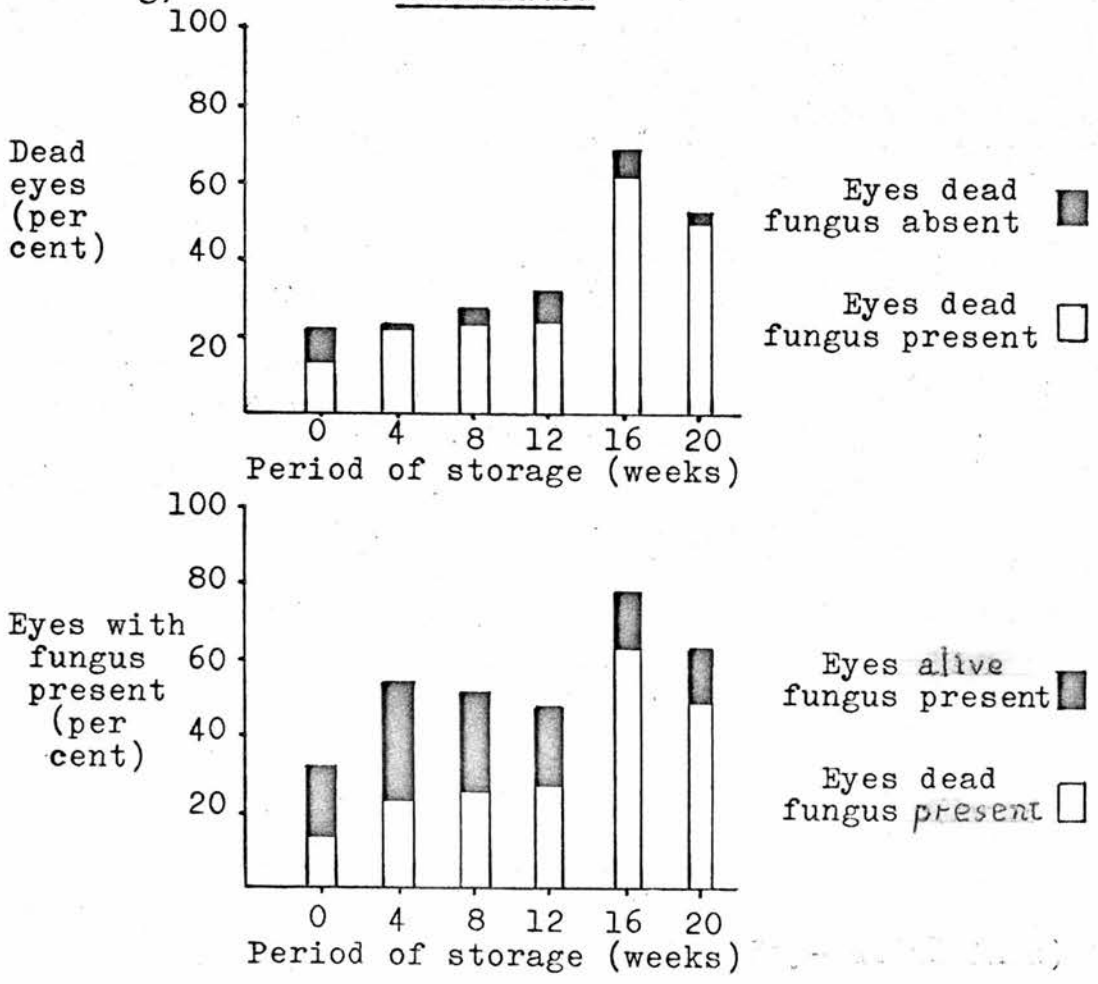
Humidity Damp



Temperature 5°C

Humidity Dry

g) Inoculated



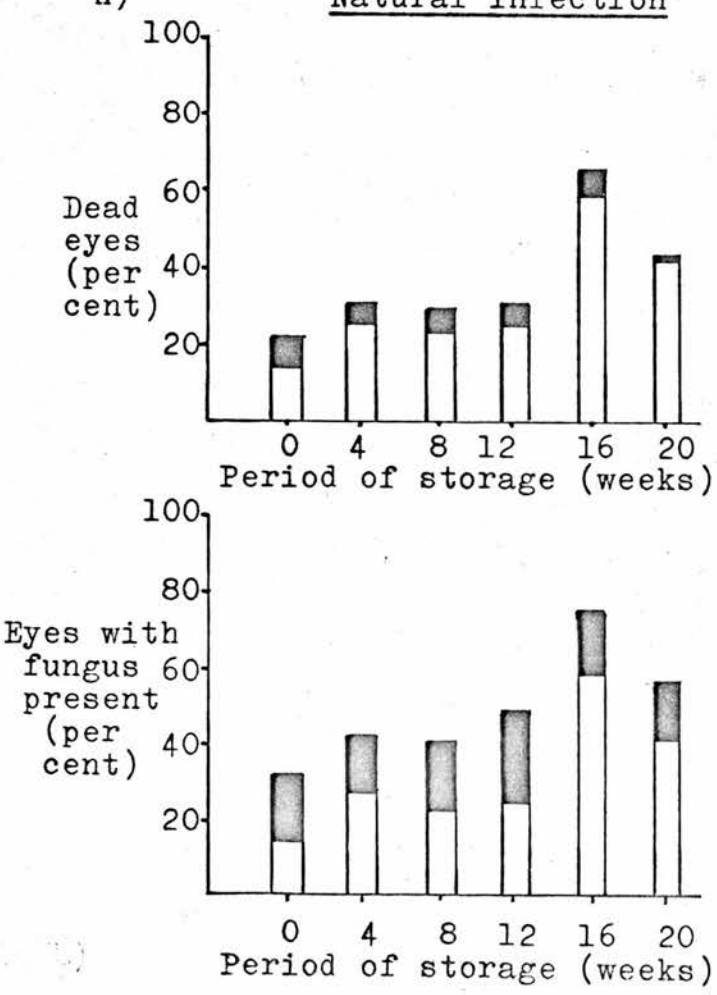
Eyes dead fungus absent

Eyes dead fungus present

Eyes alive fungus present

Eyes dead fungus present

h) Natural infection



between 12 and 16 weeks and could be accounted for by infected dead eyes. All treatments at this temperature showed a progressive increase in the level of the fungus during the storage period which was more significant than that shown in the 10°C treatments. Up until the 12-16 week period of storage the numbers of dead and live eyes infected in the various treatments was approximately equal, but after this period the dead eye figure increased to give a ratio of dead to live eyes of about 80:20. It appears that in this 12-16 week period a large number of hitherto live eyes were killed off. The effect of inoculation in storage at 5°C (Figs. 9e,g) was similar to that at 10°C in that there was an increase of infection at the 4-8 week storage compared with the corresponding natural infection treatments (Figs. 9f, h). There was some evidence that the effect of the additional inoculum increased the fungal level at the end of the storage period, this being especially true in the damp treatments (Figs. 9e, f). However, since there was no difference in either the 4-8 week or final dead eye figures for the corresponding natural infection and inoculated treatments, this increase in Oospora pustulans level by artificial inoculation was not associated with an increase in the numbers of dead eyes. From this evidence it would appear that the infection which subsequently caused eye damage must have been established in the tissue by lifting time and any subsequent infection caused little practical damage. The effect of high humidity in the 5°C treatments was mainly to magnify the basic effects occurring in

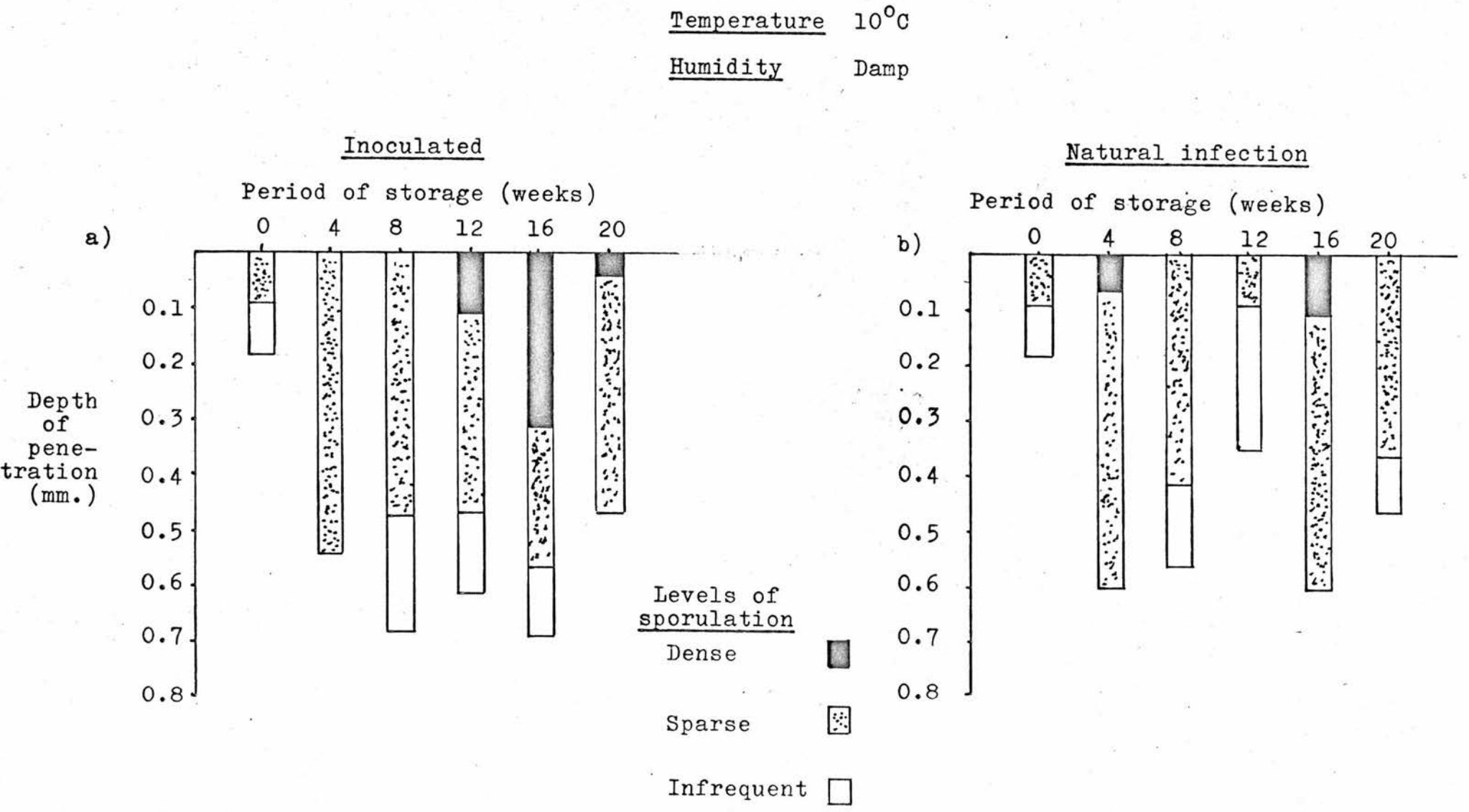
the dry treatments, i.e. an increase in dead eyes and an increase in the level of the fungus during storage. The high values of the 16 week figures for percentage of dead eyes and percentage of infected eyes for the 2 dry treatments (Figs. 9g, h) is difficult to explain especially in the light of the lower figures in the 20 week treatments.

From the evidence of this experiment it may be said that low temperature exerted the main effect in increasing the number of dead eyes infected by Oospora pustulans. High humidity served to increase the level of fungal infection, more so at 5°C than at 10°C and while at 5°C it had some effect on the number of infected dead eyes produced this was not so at 10°C.

3.2. Depth of penetration of infected eyes

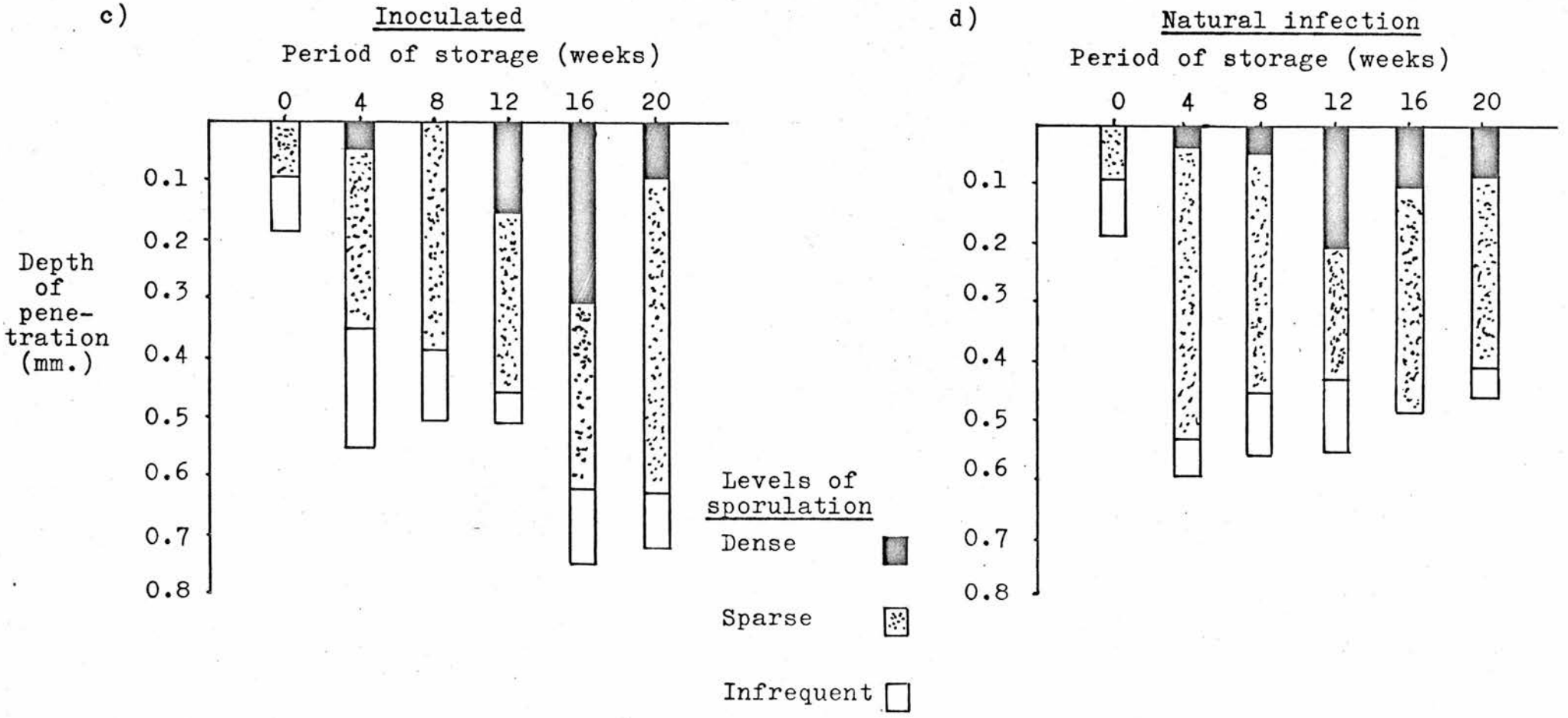
The results for the assessments of depth of penetration by the fungus (Figs. 10a-h and Appendix XXIIa, b) show that there was a marked increase in the depth of penetration between lifting time and after 4 weeks of storage. It must be borne in mind that lifting time was late, 3rd November, and thus the 4 week examination was not made until 6th December. Related to normal lifting time 3rd November would be about 3 weeks later and 6th December about 7-8 weeks. Using such time scales these results agreed reasonably well with previous work in this section showing that the depth of penetration of the fungus increased markedly between 6 and 9 weeks of clamp storage, after lifting at the normal time.

Figure 10. The average depths of the different levels of sporulation (mm.) and the total average depth of penetration of Oospora pustulans (mm.) into the tissue of tubers naturally infected and artificially inoculated and subjected to different storage humidity and temperature regimes.



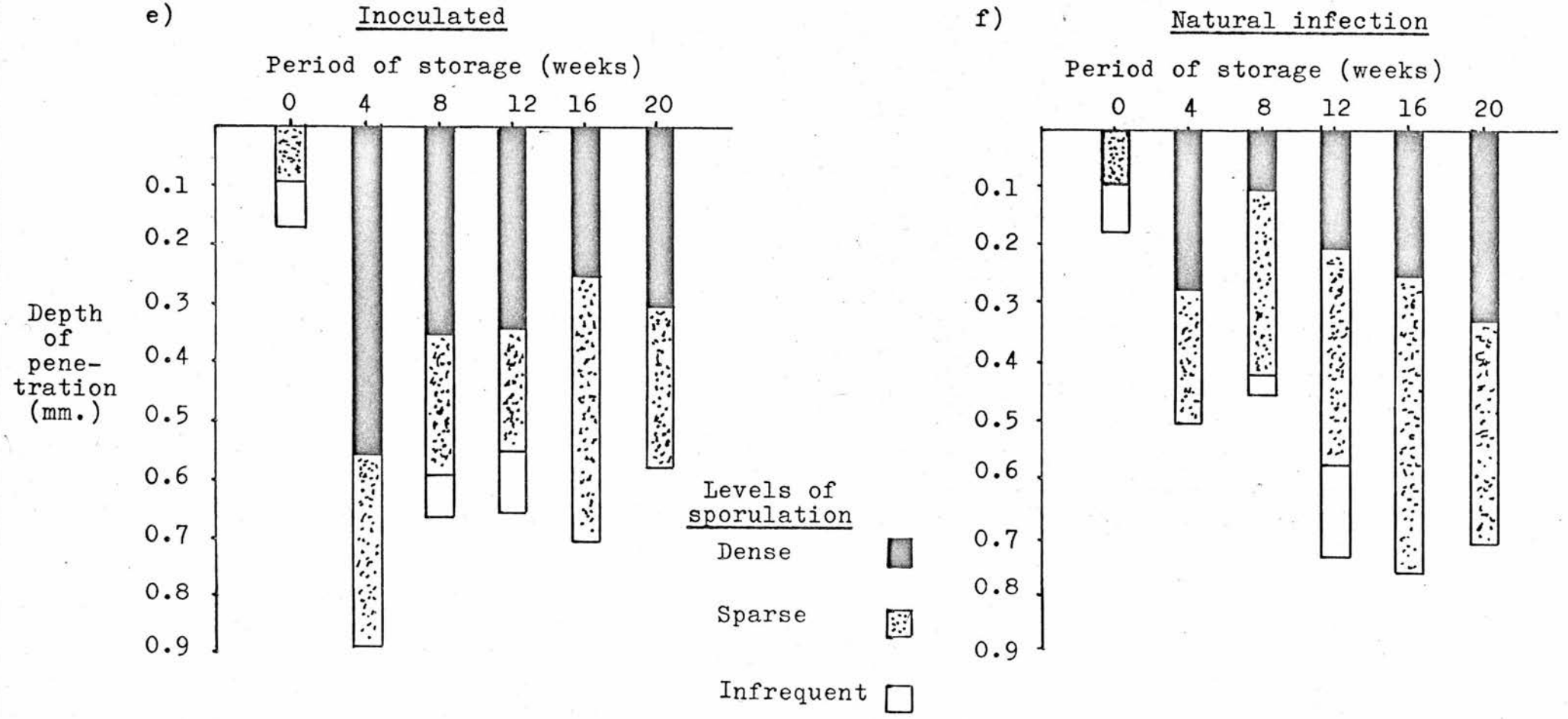
Temperature 10°C

Humidity Dry



Temperature 5°C

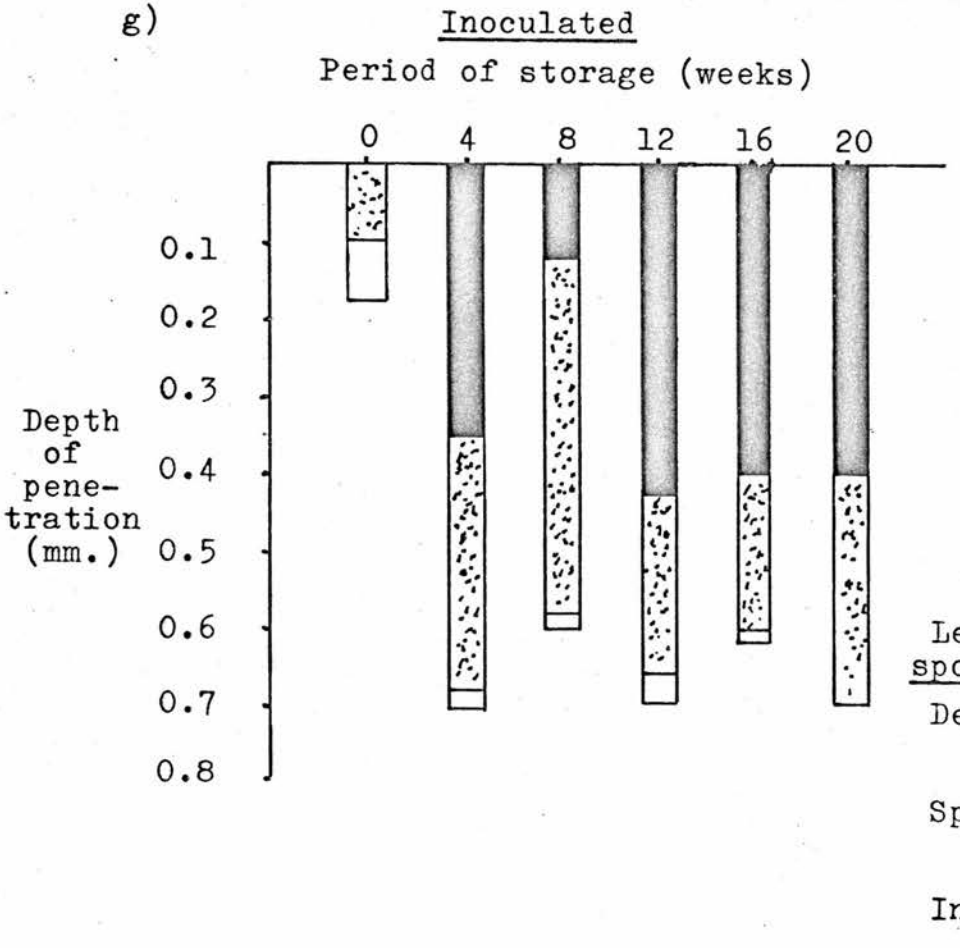
Humidity Damp



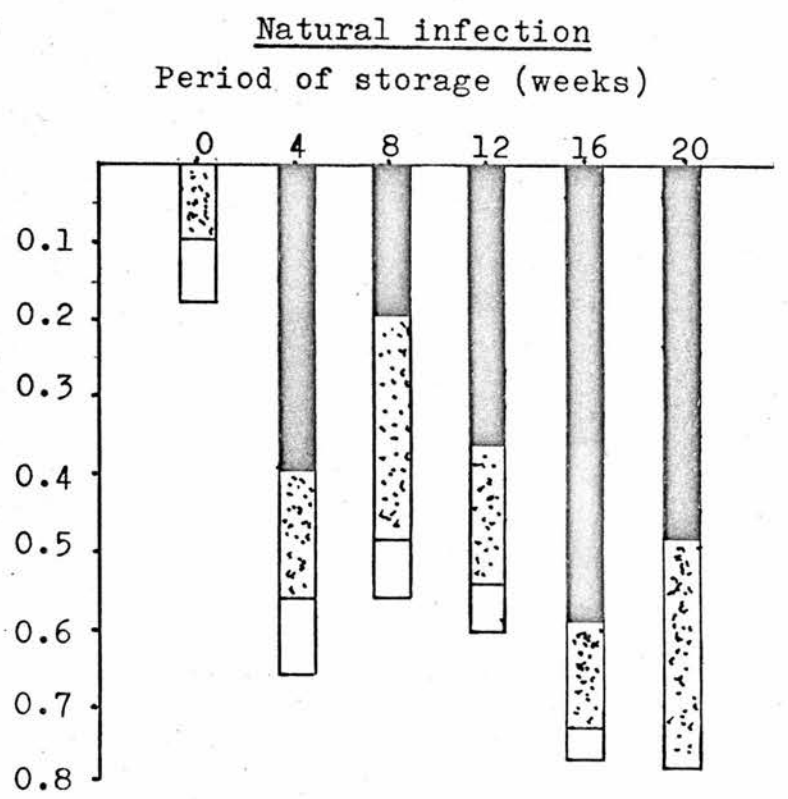
Temperature 5°C

Humidity Dry

g)



h)



Within the various treatments no definite pattern was set in terms of a progressive increase in depth of penetration as the storage season advanced. By early December the pathogen appeared to have penetrated the eye tissue to the maximum depth. There was found to be no significant differences between the levels of penetration under the two temperature regimes (Appendix XXIII), but there were differences in the densities of fungal growth in the tissue. This was probably due to the fact that growth of Oospora pustulans in an infection site was more vigorous at lower temperatures and this was reflected in pustules of greater diameter.

Level of humidity and artificial inoculation had no effect on the depth of penetration or on the density of fungal growth in the tissues. This apparent ineffectiveness of artificial inoculation lends weight to the evidence from the results of surface infection that artificial inoculation at lifting was a superficial infection causing little practical damage to the tissue. However, this may be due to the late lifting time in this experiment, since other workers have shown that artificial inoculation on tubers lifted early or at the normal time will produce normal skin spot pustules.

Discussion

The results indicate that, while time of haulm destruction had no appreciable effect on the subsequent level of skin spot infection in a crop, time of lifting had an important bearing on disease development under certain storage treatments. If tubers were lifted before mid-September and subjected to box storage and disinfection with an organo-mercury solution or box storage alone, then, compared with clamp storage, a reasonable level of control of skin spot could be achieved. As lifting time was delayed after mid-September, however, the efficiency of boxing alone, as a control measure, gradually diminished until by later lifting dates the level of disease development from this treatment was similar to that from clamp storage. Boxing and disinfecting, on the other hand, continued to give a high level of control at later lifting dates and it also proved effective where tubers were treated after lifting and clamp storage, but to a decreasing extent as the period of clamp storage was prolonged.

In the field it would appear that a progressive development of infection by the fungus occurred and that this development could be arrested in its early stage by boxing alone. The check to fungal activity by boxing might be attributed to exposure of the tubers to dry conditions, compared with damp conditions in a clamp or in the soil. In later stages of infection reasonable reduction in the disease could only be achieved by fungicidal disinfection, but the period during

which this was possible was also limited.

The failure of boxing to provide effective disease control at later dates of lifting may relate to the establishment of the pathogen in the tissues having reached a stage that it could no longer be checked by changing humidity or to lower ambient temperatures at later lifting times rendering the boxing treatment less efficient. The importance of high temperature at lifting was seen in small scale studies where tubers lifted at the normal time and subjected to continuous storage at 15°C in damp conditions developed significantly less skin spot than tubers stored under lower temperature conditions. The results of similar small scale experimental work by Lennard (1967, unpublished) are in agreement with this. In this case tubers lifted at the various times in conjunction with the field trial in 1966 were immediately stored under cold, damp or warm, dry conditions after lifting. It was found that, where a temperature of 10°C was maintained, exposure of tubers to dry conditions after lifting gave an effective control of skin spot at later dates of lifting. However, eye infection still tended to increase as lifting was delayed, this possibly relating to a more rapid establishment of eye infection in the field, as also suggested by the results of the field trial. Lennard also examined the effects of a curing period of high temperature at the beginning of storage on disease development, but the results of a small scale trial indicated that a high temperature must be coupled with a dry atmosphere or carried over a long period, if the humidity was

high, to achieve any appreciable control. In practice, however, such temperatures for long periods would induce excessive sprouting and weight loss. The findings from these small scale trials indicate that control of skin spot by boxing alone can be extended to later times of lifting, but relatively high temperatures during at least the earlier part of storage, are required. It would appear that boxing alone will be most effective if lifting is early, and thus associated with relatively high temperatures of about 10°C or that such temperatures are induced artificially if lifting is later and ambient temperatures at the time are not high enough. The loss in yield by lifting in mid-September was not found to be particularly great and moreover may be offset, to some extent, by a lower incidence of tuber blight in blight susceptible varieties where early haulm destruction is carried out.

Studies on the depth of penetration of the pathogen into the host tissue has shown that by about 6-8 weeks after normal lifting time the fungus had penetrated the tissue to the maximum depth achieved, evidence in agreement with that from the histological work of Allen (1957). This time period coincides with the approximate period up to which chemical control of the disease is still possible in tubers stored in clamps after lifting, indicating that by 6-8 weeks the fungus is established in the tissue to an extent which cannot be controlled by the fungicide. The variation in the time after lifting when chemical control could still be achieved has been associated with temperature conditions in the first 6 weeks of

storage, the control period being shorter when the temperatures were lower. No corresponding data are available concerning the depth of penetration of the fungus under varying temperatures from lifting, but where tubers were stored under different temperature and humidity conditions from lifting in early November it was found that at 5°C the growth of the fungus was more vigorous than at 15°C suggesting that with low temperatures in storage the fungus will establish itself in the tissues at an earlier stage than with higher temperatures.

The microscopic eye tests made on the tubers stored in clamps showed a higher incidence of infected eyes and a greater number of dead eyes at the end of the storage period than there was at lifting time. With tubers stored under different temperature and humidity conditions a progressive increase in the number of infection sites and number of dead eyes was found under low temperature and high humidity conditions. An apparent increase in the number of infection sites during storage was also found by Edie (1966) who suggested that the ability of the fungus to sporulate may increase at later stages of storage, but considered it to be more likely that there was a spread of infection during storage and an actual increase in the total number of infected eyes in agreement with Kharkova (1961b) who found Oospora pustulans conidia in the atmosphere of a potato store. However, while spread of the fungus may take place in storage it does not appear to account for the increase in number of infected dead eyes since artificial inoculation in early November, while increasing the incidence

of infection sites was found to have no effect in increasing the number of dead eyes. Moreover, the increase in number of infection sites from artificial inoculation was of a relatively low order which might suggest that spread of the fungus during storage does not readily become established.

The importance of humidity conditions of storage on disease development has been established. Variations in soil moisture content at the time of lifting may thus exert an effect on subsequent disease development through an influence on the humidity of the storage atmosphere. However, in investigations on the effects of varying levels of soil moisture on skin spot infection samples from the different treatments were all stored in the same clamp and thus any effect of soil moisture on storage humidity might have tended to be masked. The results, however, did suggest that skin spot infection was higher with moderate soil moisture levels than with dry soils, although very high levels of moisture also gave low levels of infection.

SECTION B

Seed tuber treatments for the control of skin spot in relation to subsequent field effects and skin spot development in the following crop.

Introduction

Experimental work, as described in Section A, has been concerned with measures taken during the storage of seed tubers which might reduce the level of skin spot development. The experimental work in this section includes an investigation of the effects of certain of these treatments on subsequent growth in terms of rate of emergence, level of blanking and yield from planting treated tubers and also on the transmission of skin spot infection to the resulting crop. Two experiments, related to Expt. A.1., were carried out. The first was concerned with planting seed tubers periodically removed from clamp storage and subjected to treatments of boxing and boxing with organo-mercury disinfection. In earlier work Edie (1964) found that such treatments had little effect on the growth of the seed, while the treatments which reduced skin spot incidence in the seed tubers gave lower levels of infection in the subsequent crop. The second experiment was related to the work where the effects of various non-mercury fungicides on skin spot development were examined. In this case further effects of these fungicides with respect to crop growth were investigated.

It has been shown that organo-mercury disinfecting of seed

tubers at planting reduces skin spot development on the resulting crop, though not to the same extent as that from disinfecting at lifting (Greeves and Muskett, 1939; Edie, 1964). It has also been reported that disinfecting at planting has an effect on the emergence rate or yield of planted seed (Edie, 1964). An experiment was therefore carried out to examine further the effects of seed disinfection at planting on field growth and disease transmission to the subsequent crop from seed showing varying degrees of skin spot infection and carrying varying levels of inoculum of Oospora pustulans as measured by incubation and microscopic examination of tuber eyes.

Skin spot infection of King Edward seed has the effect during storage of severely damaging or killing the bud tissues of the eyes resulting in delays in emergence or blanking in the field. In the final experiment of this section tubers were sprouted during the latter half of the storage period to see if such a treatment could overcome to any extent the damaging effects of skin spot infection.

Experimental Work

B.1. Effects of delayed boxing and disinfection treatments of seed tubers on plant emergence, yield and skin spot development in the subsequent crop.

1. Materials and Methods

In growing seasons 1965 and 1966 King Edward seed tubers were randomly selected from the appropriate treatments in Expt. A.1. for the previous storage season. The treatments used were as follows, having all been lifted in early October.

(a) Disinfected and boxed at lifting.

(b) Boxed at lifting.

(c) Clamped until December then boxed.

(d) Clamped throughout storage season.

The treatments were planted in plots replicated 4 times in a randomised block lay-out. In 1965 there were 15 tubers per plot and in 1966 there were 12. The tubers were planted in a medium-loam soil on 22nd April 1965 and 18th April 1966. Periodic emergence counts were taken during the growing season. The plots were lifted on 12th October, 1965, and 15th October, 1966, when yield assessments were made, the tubers placed in net bags and stored in a clamp until the following March when they were examined for skin spot development.

2. Results

The results of the assessments of rate of emergence,

blanking and yield in relation to the treatments are recorded on Table 12 and Appendix XXIV and those for skin spot development on Table 13 and Appendix XXV.

2.1. Emergence

No blanking was found in the plots of seed tubers, boxed and disinfected at lifting and the higher levels of blanking were associated with either delayed boxing from clamp storage or continuous clamp storage.

Table 12. Effect of delayed boxing and disinfection treatments on rate of plant emergence, blanking and yield of the subsequent crop in growing seasons 1965 and 1966.

Treatment	Average number of days to emergence		Blanking (per cent)		Average yield per plot (lbs.)	
	1965	1966	1965	1966	1965	1966
Disinfected and boxed at lifting	52.2	50.3	0	0	17.3	25.5
Boxed at lifting	54.2	52.1	2	4.2	16.6	26.0
Clamped until December	59.2	54.3	5	10.5	15.8	22.0
Clamped continuously	54.8	54.7	13	2.1	17.5	24.0

There was, however, no significant effect of the various storage treatments on emergence rate of plants that did emerge (Appendix XXIVa, b).

2.2. Yield

In both years plot yields were very variable and there were no significant differences between the treatments (Appendix XXIVc, d). Edie (1964) in similar findings suggested that the original infection levels of the treated seed tubers, which ranged from 0.72 to 8.6 surface infection index, were not high enough to reflect any differences from the treatments. However, the infection levels on the planted seed in 1965 ranged from 2.1 to 14.5 (Table 13) and it may be concluded that these treatments had no effect on yield even where substantial levels of infection on the original seed tubers were involved.

2.3. Skin Spot Development

Table 13 shows the level of skin spot on the planted seed and that which developed on the resulting crop. Comparing the various treatments the skin spot development showed a similar pattern in both years. Only the disinfection and boxing treatment gave any significant reduction in skin spot in the harvested crop (Appendix XXV). This was especially well demonstrated with eye infection in both years. Differences in levels of surface infection were not significant in 1965, but they did follow the same trend as in 1966 where boxing and disinfection gave significantly less infection than the other treatments. While boxing at lifting gave some reduction in level of surface infection on the planted seed it gave no apparent reduction in level of eye infection compared

Table 13. Effect of delayed boxing and disinfection treatments on the skin spot level on the planted seed and the disease development in the harvested crop in seasons 1965 and 1966.

Treatment	Planted seed				Harvested crop			
	S.I.I.		E.I.I.		S.I.I.		E.I.I.	
	1965	1966	1965	1966	1965	1966	1965	1966
Disinfected and boxed at lifting	2.1	0.7	18.0	13.0	5.6	4.4	37.5	18.0
Boxed at lifting	2.1	3.7	58.0	43.0	9.4	8.5	46.1	39.7
Clamped until December	11.6	6.7	58.0	37.0	11.0	9.3	52.4	39.2
Clamped continuously	14.5	6.2	56.0	42.0	5.3	9.4	57.0	44.0

with that from clamp storage and levels of infection on the resulting crop were similar to those of seed tubers from clamp storage.

In comparing the responses between the 2 years, the level of surface infection on the planted seed in 1966 was low compared with that in 1965 but was almost as high on the harvested seed in 1966 as in 1965. However, the degrees of eye infection on the planted and harvested seed in 1966 were both lower than the corresponding figures in 1965 suggesting that the carry-over effect of the seed inoculum was more marked in the case of eye infection than in the case of surface infection.

B.2. Effects of disinfection treatment of seed tubers with different fungicides at lifting time on plant emergence, yield and skin spot development on the subsequent crop.

1. Materials and Methods

In growing season 1966 King Edward seed tubers were randomly selected from 3 treatments in Expt. A.1. for the previous storage season, as follows:

(a) Lifted 6 October 1965, disinfected e.e.m.c.λ ½ lb in 10 gal 3 min and boxed;

(b) Lifted 6 October 1965, disinfected Maneb λ (Manganese ethylene bisdithiocarbamate) (80 per cent DP) ¾ lb in 10 gal 3 min. and boxed;

(c) Lifted 6 October 1965, boxed only.

The treatments were carried out immediately after lifting and the tubers then stored in an insulated shed until March 1966 when they were assessed for skin spot development.

The treatments were planted on 15th April 1966 on a medium-loam soil in plots replicated 4 times in a randomised block lay-out with 15 tubers per plot. Periodic emergence counts were made during the growing season. The plots were lifted on 15th October 1966, when yield assessments were made and the tubers stored in net bags in a clamp until the following March when the treatments were assessed for skin spot development.

2. Results

There was no significant difference between the emergence

rates from the different treatments which all gave complete plant stands (Table 14 and Appendix XXVIa). The results for yield also showed no significant differences between treatments (Appendix XXVIb).

Table 14. Effects of disinfection treatments of seed tubers with different fungicides at lifting time on rate of plant emergence, yield and skin spot development in the resulting crop.

Field and disease factors	Treatment		
	Disinfected with e.e.m.c.	Disinfected with Maneb	Boxed only
Average number of days to emergence	57.7	55.3	58.6
Average yield per plot (lbs.)	19.0	23.0	21.0
S.I.I.	3.8	6.3	6.5
E.I.I.	15.8	28.0	27.9

However, the assessments of skin spot infection on the harvested crops showed that disease was less from the e.e.m.c. disinfected treatment than from Maneb or boxing only treatments, the reduction being significant in the case of eye infection (Appendix XXVIc, d). Although Maneb gave a reduced level of infection on the planted seed compared with boxing alone there were no differences in levels of infection on the resulting crop between these treatments.

B.3. The effect of disinfection of seed tubers at planting on plant emergence and the transmission of skin spot infection to the resulting crop.

1. Materials and Methods

King Edward seed tubers of two different levels of infection were disinfected with e.e.m.c. solution one day before planting. Four treatments were used in planting:

(a) Tubers free from obvious skin and eye infection and disinfected;

(b) As in (a) but not disinfected;

(c) Tubers with slight surface infection, some eye infection and disinfected;

(d) As in (c) but not disinfected.

At the time of planting 20 tuber samples were taken from each of these treatments for microscopic eye tests. The planting date was 16th April 1964 on a medium-loam soil in plots replicated 4 times for each treatment in a randomised block lay-out with 7 tubers per plot. Periodic emergence counts were taken during the growing season, the plots were lifted on 15th October 1964 and the tubers stored in net bags in a clamp until March 1965 when assessments of skin spot development were made.

2. Results

2.1. Microscopic Eye Tests

In the tubers visibly free of skin spot (Table 15) which

had not been disinfected it may be seen that 13% of the eyes supported growth of the fungus whereas no fungus was detected in comparable tubers which had been disinfected.

Table 15. Effects of disinfection with e.e.m.c. on the level of Oospora pustulans present and the viability of eyes on tubers with different levels of skin spot infection as examined by microscopic eye tests.

Disease level on planted seed	Treatment	Eyes alive (per cent)		Eyes dead (per cent)	
		Fungus present	Fungus absent	Fungus present	Fungus absent
Slight surface - some eye infection	Not disinfected	24.3	35.4	29.6	10.5
	Disinfected with e.e.m.c.	5.1	56.9	0	38.0
Free from obvious infection on surface or eyes	Not disinfected	13.1	77.4	0	9.5
	Disinfected with e.e.m.c.	0	91.6	0	8.4

The disinfection treatment also decreased very considerably the number of eyes of visibly infected tubers on which the fungus was found.

2.2. Emergence

These results (Table 16 and Appendix XXVIIa) showed there to be a significantly slower emergence rate from visibly infected seed compared with seed 'free' from symptoms and from

disinfected compared with non-disinfected tubers.

Table 16. Effects of disinfection at planting of tubers with different levels of skin spot infection on plant emergence and skin spot development in the resulting crop.

Disease level on planted seed	Treatment	Average number of days emergence	Skin spot development	
			S.I.I.	E.I.I.
Slight surface, some eye infection	Not disinfected	44.8	9.6	66.7
	Disinfected with e.e.m.c.	46.7	5.8	43.0
Free from obvious infection on surface or eyes	Not disinfected	42.3	7.8	44.5
	Disinfected with e.e.m.c.	45.2	4.5	23.3

The delay associated with visibly infected seed may be attributed to bud death in infected eyes and that caused by disinfecting tubers to a phytotoxic effect of mercury on the sprouts.

2.3. Skin Spot Development

There was seen to be significantly less surface and eye infection (Table 16 and Appendix XXVIIb, c) for the disinfected compared with non-disinfected treatments suggesting that the inoculum must have been considerably reduced by the disinfecting process. The 'free' from symptoms' seed was seen to produce infection in the resulting crop despite the fact that no skin spot pustules were observed on the seed nor was there any fungus

capable of sporulating on the surface of the tubers at planting. To what extent this was due to possible soil contamination or seed contamination is not known. The visibly infected seed category gave a higher level of infection in the resulting crop than the 'free' category. This was significant in the case of eye infection, but, although the same trend was there, there was no significant difference between the treatments in surface infection.

B.4. Effects of sprouting tubers at different temperatures on plant emergence, yield and skin spot development in the subsequent crop.

1. Materials and Methods

King Edward seed tubers were removed from clamp storage on 9th February 1966 and placed in chitting trays holding about 35 lbs. of tubers in each. These tubers were then sprouted under two sets of conditions:

(a) Stored at about 10°C under conditions of high humidity;

(b) Stored at about 15°C in dry conditions.

After 3 weeks in these environments the boxes from both treatments were held in an insulated shed subject to fluctuating temperatures (6°C - 9°C). Three planting treatments were used consisting of samples randomly selected from tubers subjected to each of the above two sprouting treatments and tubers which had been kept in continual clamp storage. The condition of the sprouts at planting time in the various treatments were as follows:

(a) 10°C - humid - sprouts average length 3", long and thin;

(b) 15°C - dry - sprouts average length 1¼", shorter and thicker than in (a);

(c) Clamp storage - sprouts just beginning to emerge from the eye.

The tubers were planted on 18th April 1966 in a medium-loam soil in plots replicated 4 times for each treatment in a

randomised block lay-out with 12 tubers per plot. Periodic emergence counts were taken during the growing season. The plots were lifted on 15th October 1966, yield assessments made and the tubers stored in net bags in a clamp until March 1967 when assessments on the skin spot development were made.

2. Results

The results for rate of plant emergence, yield and skin spot development on the resulting crops are shown on Table 17 and Appendix XXVIII. All treatments gave complete plant stands, but the number of days to emergence was significantly less for the sprouted treatments than for the non-sprouted treatment.

Table 17. Effects of sprouting tubers at different temperatures on plant emergence, yield and skin spot development in the resulting crop.

Sprouting treatment	Average number of days to emergence	Average yield/plot (lbs.)	Skin spot development	
			S.I.I.	E.I.I.
Sprouted at 15°C	40.2	36.0	13.9	57.3
Sprouted at 10°C	41.7	36.8	13.0	53.4
Unsprouted	52.3	33.0	13.0	45.7

The yield data, however, showed no significant differences between treatments and assessments of skin spot infection on

the resulting crops indicated no significant differences between treatments in their effects on seed borne transmission of the disease.

Discussion

The experimental work, comparing the effects on disease development of fungicidal treatments at lifting with boxing alone at lifting and clamp storage treatments, established that organo-mercury disinfection markedly reduced the level of skin spot development on the seed compared with all the other treatments, while boxing alone and boxing with Maneb disinfection gave some reduction in comparison with the clamped treatments. However, although seed tubers from boxing and disinfecting or boxing only at lifting treatments gave less blanking in the field than those from clamp storage treatments, there were no differences in the emergence rates of plants which did emerge or in the final yields between the treatments. In treatments where tubers were disinfected with an organo-mercury compound at lifting there was significantly less skin spot on the resulting crop compared with the other treatments, no doubt a reflection of low level of inoculum on the organo-mercury disinfected planted seed compared with the other planted seed.

Disinfecting seed at planting time with organo-mercury was seen to reduce the fungal inoculum on the seed, as measured by microscopic eye tests, and this was reflected in a significant reduction of skin spot infection in the resulting crop compared with that of non-disinfected seed tubers. Disinfection at planting was found to delay emergence of the planted seed and this may be attributed to a phytotoxic effect of the mercury on the sprouts. However, the results from

microscopic eye tests indicated no difference in numbers of viable eyes between disinfected and non-disinfected tubers, suggesting that none of the eyes were completely killed by the disinfection treatment and it was noted that this treatment did not give rise to blanking. The presence of infected eyes in the visibly infected seed category resulted in a delay in emergence compared with 'free' of symptoms seed, but the seed inoculum difference was not reflected in the disease levels of the resulting crops.

While tubers sprouted late in the storage season showed an increased rate of plant emergence compared with that from seed kept in continuous clamp storage, no differences were observed in the level of blanking or in the yield between the treatments. The levels of seed inoculum were more or less equal for the 3 treatments and the levels of skin spot infection on the resulting crop were similar. The eye infection (E.I.I. = 41.7) on the planted seed which had not been sprouted may have been too low to give a reasonable level of blanking. **This could have been used to measure the success of sprouting in overcoming the damaging effect of skin spot infection.**

SECTION C

Field responses to varying levels of skin spot infection on seed tubers of different varieties planted in different types of soil.

Introduction

The effects of varying levels of skin spot infection on seed tubers on the rate of emergence, amount of blanking and yield and on the transmission of disease to the resulting crop has been investigated thoroughly in the variety King Edward (Boyd and Lennard, 1961a). This work, however, has never been carried out to a comparable extent on other varieties and, as Nagdy and Boyd (1965) suggest, varietal characteristics may be of such a nature as to be able to overcome the damaging effects of skin spot infection of seed tubers on crop growth.

There is evidence to show that soil type has some effect on the skin spot infection of potatoes (Khrobrykh, 1959; Gomolyako, 1959; Salt, 1964), but there is no report of the effect of soil type on emergence rate, incidence of blanking and yield when infected seed is planted.

The first part of the experimental work in this section was set out to examine in detail the effects of Oospora pustulans infection on sprout growth of seed tubers of Kerrs Pink, a vigorous sprouting variety, and King Edward, a variety showing less vigorous sprout growth. Both varieties are recognised as being highly susceptible to skin spot infection,

but King Edward would appear from field experience to be much more prone to delayed emergence and blanking due to the disease. It was hoped that this difference in sprouting vigour could be related to the field responses of these varieties as examined in the second part of the experimental work, which was also concerned with gaining more information on the combined effects of soil type, variety and level of seed inoculum on crop growth and transmission of disease to the resulting crops.

Eye necrosis was caused largely by O. p in artificial inoculations but can therefore conclude that necrosis in Kerr's Pink does not markedly inhibit sprout development. Same argument can be applied to King Edward in spite of the original level of necrosis of doubtful origin. But what is the reason?

EXPERIMENTAL WORK

C.1. A comparison of the effects of natural and artificial infection of Oospora pustulans on sprouted tubers of the varieties King Edward and Kerrs Pink.

1. Materials and Methods

The tubers used were samples of the varieties King Edward and Kerrs Pink which had been washed, surface sterilised with formalin and kept in chitting trays since lifting time in early October 1964. Twenty tubers were taken for each variety, placed 5 per box in damp cardboard boxes and kept in an incubator at 15°C for 14 days as from 4th November 1964. The tubers were then removed and all the eyes of each tuber examined for necrosis of the bud tissues. The tubers of each variety were then divided into 2 lots of equal number and 1 lot artificially inoculated with a spore suspension of Oospora pustulans (10^6 spores/ml. for 1 min.) and stored in darkness in damp cardboard boxes at approximately 5°C until May 1965. The second lot of each variety was dipped in sterile water for 1 min. and stored in the same manner.

On 14th May 1965 the tuber eyes were again examined and assessed for bud necrosis and Oospora pustulans sporulation. The tubers were then placed in chitting trays and allowed to sprout in a potato store until 26th July 1965 when a further examination of the eyes was carried out recording the extent of active sprout growth related to the eye condition as of 14th May 1965.

2. Results

Table 18 summarises the eye condition of the varieties before and after winter storage. Before storage at 5°C, 39 per cent of the eyes of the King Edward tubers were necrotic as compared with 25 per cent for Kerrs Pink tubers. In the uninoculated treatment, after storage at 5°C, the King Edward eye necrosis more than doubled whereas in Kerrs Pink the increase in necrotic eyes was only slight.

Table 18. Level of Oospora pustulans sporulation and necrosis in eyes of inoculated and uninoculated Kerrs Pink and King Edward tubers before (Oct. 1964) and after (May 1965) storage at 5°C.

Variety	Eye necrosis before storage (per cent)	Eye necrosis and fungal sporulation after storage (per cent)			
		Tubers uninoculated		Tubers inoculated	
		Eye necrosis	Fungal sporulation	Eye necrosis	Fungal sporulation
King Edward	39	89	4	96	77
Kerrs Pink	25	32	16	79	56

The number of eyes naturally infected with Oospora pustulans was quite low being less in King Edward than Kerrs Pink, and skin spot infection could not account for the high level of eye necrosis in King Edward. As no other pathogen was evident it is suggested that the high incidence of necrosis in King Edward related to physiological damage possibly associated

with exposure of the tubers to low temperatures after active bud growth had begun and that Kerrs Pink sprouts appeared to be able to withstand such low temperatures better than King Edward. In the inoculated treatments there was almost complete eye necrosis in King Edward and a high level of fungal sporulation. Kerrs Pink also had a high level of eye necrosis and a high level of fungal sporulation although lower than the corresponding King Edward levels. In the variety Kerrs Pink the increase in eye necrosis between the uninoculated and inoculated treatments of 47 per cent was accounted for by an increase in Oospora pustulans infection of the eyes, of 40 per cent. In the variety King Edward the increase in eye necrosis was only 7 per cent associated with an increase in fungal sporulation of 73 per cent. However, it would appear that the damaging effects of the fungus on the sprouts were masked by the possible low temperature sprout damage in this variety.

Table 19 shows the incidence of active sprout growth in the various treatments in July 1965 from the total number of eyes and from the necrotic and non-necrotic eyes as recorded in May 1965. A greater number of eyes showing active sprout growth were found in Kerrs Pink than in King Edward for both the inoculated and uninoculated treatments and although artificial infection by Oospora pustulans decreased the level of sprouting in both varieties the reduction was only slight in Kerrs Pink. The necrotic eyes in King Edward generally failed to sprout whereas with Kerrs Pink the necrosis in the eyes did not seem to have any permanent damaging effect on the sprouting

Table 19. Level of active sprout growth in July 1965 from necrotic and non-necrotic eyes, as recorded in May 1965, of inoculated and uninoculated Kerrs Pink and King Edward tubers.

Variety	Active sprout growth from eyes of uninoculated tubers (per cent)			Active sprout growth from eyes of inoculated tubers (per cent)		
	Necrotic eyes	Non-necrotic eyes	Total eyes	Necrotic eyes	Non-necrotic eyes	Total eyes
King Edward	0	60	16	4	50	6
Kerrs Pink	55	61	38	29	33	30

potentialities, since there was little difference in sprout development in necrotic and non-necrotic eyes.

These results would seem to indicate that the eye necrosis in Kerrs Pink caused by Oospora pustulans infection does not markedly inhibit sprout development. In the variety King Edward, however, eye necrosis inhibits sprout development almost completely, although it is not possible to explain such eye necrosis as being caused solely by Oospora pustulans infection due to possible low temperature damage to sprouts of this variety. However, the sprout vigour of Kerrs Pink has been demonstrated as being able to overcome much of the damaging effect of Oospora pustulans infection.

C.2. Field responses to varying levels of skin spot infection on seed tubers of different varieties planted on different types of soil.

1. Materials and Methods

In 3 consecutive years, 1964-66, tubers with varying levels of skin spot infection were planted in 2 types of soil, a medium-loam and a light sandy soil. Details of the varieties planted, seed infection categories, planting treatments and dates of plot lay-outs are given in Table 20.

Emergence counts were taken at weekly intervals during the growing season. The plots were lifted in the second week of October of each year, stored in net bags in the same clamp until the following March when assessments of skin spot developments in the treatments were made. In 1965 yield assessments were made at the time of lifting.

Statistical analyses of the results was carried out by the comparison of means using Students "t" test or using analyses of variance when applicable (Appendices XXIX - XXXI).

The results are considered under three headings referring to the experiments carried out in each year.

1. The effects of varying levels of skin spot infection of King Edward seed tubers, time of planting and soil type on plant emergence and disease transmission to the resulting crop - 1964.
2. The effects of varying levels of skin spot infection of seed tubers of 2 different varieties and soil type on plant

Table 20. Experimental plan of the investigation of the field responses to varying levels of skin spot infection on seed tubers of different varieties planted on different soil types.

Year	Variety	Disease levels on planted seed	Planting Details	
			Medium-loam soil	Light sandy soil
1. 1964	King Edward	1. Free from obvious infection on skin or eyes 2. Slight surface, no eye infection 3. Slight surface, some eye infection 4. Moderate or severe surface infection, all eyes infected	Planting Treatments: 2 series of treatments planted at a 2 week interval, second series held in chitting trays during interval Planting Dates: 15 and 30 April Lay-out: Both series in the same randomised block, times 4 replication, 15 tubers per plot	Planting Treatments: 1 series of treatments planted Planting Date: 15 April Lay-out: Unreplicated plots, 20 tubers per plot
2. 1965	King Edward Kerrs Pink	1. Free from obvious infection on skin or eyes 2. Slight surface, no eye infection 3. Slight surface, some eye infection 4. Severe surface infection, all eyes infected	Planting Treatments: 1 series of treatments per variety planted Planting Date: 22 April Lay-out: Each variety in separate randomised blocks, times 4 replication, 15 tubers per plot	Planting Treatments: 1 series of treatments for King Edward variety planted Planting Date: 14 April Lay-out: Randomised block, times 4 replication, 10 tubers per plot
3. 1966	King Edward Kerrs Pink Majestic Redskin	1. Trace or free surface infection, no eye infection 2. Severe or moderate surface infection, all eyes infected	Planting Treatments: 1 series of treatments per variety planted Planting Date: 18 April Lay-out: All varieties in same randomised block, times 4 replication, 12 tubers per plot	Duplication of treatments and lay-out in medium-loam soil Planting Date: 19 April

emergence, yield and disease transmission to the resulting crop - 1965.

3. The effects of varying levels of skin spot infection of seed tubers of different varieties and soil type on plant emergence and disease transmission to the resulting crop - 1966.

2. Results

2.1. The effects of varying levels of skin spot infection of King Edward seed tubers, time of planting and soil type on plant emergence and disease transmission to the resulting crop - 1964.

2.1.1. Emergence

The rate of emergence and the incidence of blanking in relation to the various treatments are shown on Table 21. The rate of emergence figure for the later planted seed refers to the average number of days since the first planting date, i.e. 15th April. These results showed that tubers held in chitting trays for 2 weeks before planting emerged at about the same time as those planted 2 weeks earlier without chitting (Appendix XXIXa). On the medium-loam soil severe or moderately infected seed had a significantly slower rate of emergence than that from the other infection categories (all of) which showed similar rates of emergence. In the light sandy soil, however, there were no differences in the rate of emergence between any of the disease categories. In general, the rate

Table 21. The effect on plant emergence of different levels of skin spot infection on King Edward seed tubers planted on 2 dates and on 2 soil types - 1964.

Soil Type	Seed Infection	Planted 16 April 1964		Planted 30 April 1964	
		Average number of days to emergence	Blanking (per cent)	Average number of days to emergence from 16 April	Blanking (per cent)
Med-loam	Mod. or sev., all eyes	63.8	88.3	68.2	75.0
	Slight, some eyes	48.3	0	48.2	8.3
	Slight, no eyes	51.3	11.5	53.8	11.6
	Free	45.4	0	48.3	1.6
Sandy	Mod. or sev., all eyes	34.0	20.0	-	-
	Slight, some eyes	36.1	0	-	-
	Slight, no eyes	33.5	0	-	-
	Free	36.5	5.0	-	-

of emergence was more rapid on the light sandy soil compared with that on the medium-loam soil (Appendix XXIXa, b).

Blanking occurred in both soil types, but was more extensive in the medium-loam soil. It was also apparent that the level of blanking was greatest in the severe or moderately infected seed.

2.1.2. Skin Spot Development

Table 22 shows the level of skin spot development on the subsequent crop from the various treatments. It may be seen that the time of planting of seed tubers had no effect on the skin spot development on the resulting crop (Appendix XXIXc, e). There was, however, a definite difference in the overall infection from the seed grown on the medium-loam soil and that grown from the light sandy soil.

Table 22. The effects on skin spot development in the resulting crop of different levels of skin spot infection on King Edward seed tubers planted on 2 dates and on 2 soil types - 1964.

Soil Type	Seed Infection	Planted 16 April 1964		Planted 30 April 1964	
		S.I.I.	E.I.I.	S.I.I.	E.I.I.
Med-loam	Mod. or sev., all eyes	8.6	67.5	12.9	76.1
	Slight, some eyes	11.9	75.5	15.8	75.1
	Slight, no eyes	11.3	71.8	10.9	75.0
	Free	7.7	47.7	6.8	45.8
Sandy	Mod. or sev., all eyes	7.7	52.5	-	-
	Slight, some eyes	1.8	10.0	-	-
	Slight, no eyes	1.4	8.6	-	-
	Free	1.5	11.8	-	-

Only the tubers from the severe or moderately infected seed showed much infection in the light sandy soil and this was

only about the level of that on the tubers from the 'free' seed planted on the medium-loam soil. On the medium-loam soil the resulting crop from the 'free' seed had significantly less skin spot than that from the other seed categories which all had similar levels of infection (Appendix XXIXd, e). The low figure for the crop from the severe or moderately infected seed is difficult to explain, but it is an effect which other workers have also noted (Boyd and Lennard, 1961a; Edie, 1964).

2.2. The effects of varying levels of skin spot infection of seed tubers of 2 different varieties and soil type on plant emergence, yield and disease transmission to the resulting crop - 1965.

2.2.1. Emergence

The results (Table 23) indicate that except for the 'free' seed category the emergence rate was more rapid on the light sandy soil than on the medium-loam soil (Appendix XXXa). It was also apparent that King Edward seed showed a significantly slower rate of emergence than did Kerrs Pink seed (Appendix XXXb). In each variety the severely infected seed emerged at a significantly slower rate than the seed of the other disease categories and the average number of days to emergence increased progressively as the level of infection on the seed became greater when planted in the medium-loam soil. However, on the King Edward seed planted on the light sandy soil there was no significant difference in emergence rates between seed with the

Table 23. The effects on plant emergence and yield of different levels of skin spot infection on seed tubers of 2 varieties planted on 2 soil types - 1965.

Soil Type	Seed Infection	King Edward			Kerrs Pink		
		Average number of days to emergence	Blanking (per cent)	Total yield (lbs.)	Average number of days to emergence	Blanking (per cent)	Total yield (lbs.)
Med-loam	Severe, all eyes	66.2	80.0	22.5	60.8	27.3	60.0
	Slight, some eyes	60.3	8.3	70.5	54.8	5.0	63.0
	Slight, no eyes	55.8	15.0	61.0	48.3	1.7	65.5
	Free	54.1	8.3	62.0	47.3	1.7	66.5
Sandy	Severe, all eyes	53.8	80.0	34.5	-	-	-
	Slight, some eyes	50.6	0	62.0	-	-	-
	Slight, no eyes	47.8	0	68.0	-	-	-
	Free	51.7	0	63.5	-	-	-

various levels of infection (Appendix XXXa, b).

In general, the level of blanking in King Edward was higher than in Kerrs Pink for corresponding seed infection categories. In both varieties the level was found to be greatest from severely infected seed. In the light sandy soil the blanking of the severely infected seed in King Edward was as high as that in the medium-loam soil, but in the remaining infection

categories it was low compared to that in the medium-loam soil. Apart from the King Edward seed in the severely infected categories which gave very low yields, there were no marked differences in yield results from the different treatments. The low yield values for severely infected King Edward seed were associated with a plant stand of 20 per cent.

2.2.2. Skin Spot Development

The results are recorded on Table 24 and conclusions from statistical analysis drawn from Appendices XXIXc-e and XXXc-e. It may be seen that there was a generally higher level of infection in the King Edward seed grown on the medium-loam soil compared with that grown on the light sandy soil. As in the previous experiment there was, however, an unexpectedly low level of infection of the crop from the severely infected King Edward seed. Between the 2 varieties there was found to be no significant differences in the infection levels of the seed from the various disease categories, except for the severely infected seed where Kerrs Pink gave a higher level of infection than King Edward. On the medium-loam soil King Edward 'free' seed produced significantly less skin spot in the resulting crop than the other disease categories, except for the severe seed. On the light sandy soil, however, no significant differences were apparent between any of the tubers from the various seed infection categories. Severely infected Kerrs Pink seed gave a significantly higher surface infection index in the resulting crop than the other disease categories,

but eye infection index differences were not significant.

Table 24. The effects on skin spot development in the resulting crop of different levels of skin spot infection on seed tubers of 2 varieties planted on 2 soil types - 1965.

Soil Type	Seed Infection	King Edward		Kerrs Pink	
		S.I.I.	E.I.I.	S.I.I.	E.I.I.
Med-loam	Severe, all eyes	3.9	32.2	21.3	46.7
	Slight, some eyes	10.5	50.2	12.7	48.9
	Slight, no eyes	13.9	56.8	19.3	42.3
	Free	6.5	39.7	9.7	35.7
Sandy	Severe, all eyes	4.7	25.0	-	-
	Slight, some eyes	4.5	23.6	-	-
	Slight, no eyes	7.2	19.8	-	-
	Free	3.0	15.8	-	-

2.3. The effects of varying levels of skin spot infection of seed tubers of different varieties and soil type on plant emergence and disease transmission to the resulting crop - 1966.

2.3.1. Emergence

These results (Table 25 and Appendix XXXIa) indicate that planting seed with a high level of skin spot infection for all the varieties and in both soil types significantly delayed emergence compared with planting seed of low infection levels.

However, the effect of severely infected seed in delaying emergence was significantly greater with King Edward than it was with the other varieties. In general Kerrs Pink seed emerged at a significantly faster rate than any of the other varieties which all emerged at about the same time. It was also found that on the light sandy soil emergence was significantly more rapid than on the medium loam soil, but there were no significant interactions between the effects of soil type and varieties on the effects of soil type and the disease level of the planted seed on the emergence rate.

Kerrs Pink showed virtually no blanking at all, while in the severely infected categories of the other varieties blanking was high. With Majestic and Redskin the level of blanking was higher on the medium-loam than on the light sandy soil. With King Edward, however, the level was about the same in both types of soil. It was also much lower in the medium-loam soil than it was in the previous experiments for this variety.

2.3.2. Skin Spot Development

As is shown on Table 26 and Appendix XXXIb, severely infected seed tubers produced significantly greater levels of skin spot on the resulting crop than did seed with a lower level of skin spot for all varieties and on both soil types. However, it was found that in the variety Redskin there was a markedly greater difference in the disease levels of the crops from the 2 seed categories than there was from the other

varieties. No significant differences between the varieties in the level of skin spot carried over to the succeeding crop occurred. However, a significantly lower level of skin spot developed on tubers grown on the light sandy soil compared with those grown on the medium-loam soil. No significant interactions between soil type and variety and soil type and disease level of the planted seed in their effects on the transmission of disease to the resulting crop were found.

Table 25. The effects on plant emergence of different levels of skin spot infection on seed tubers of 4 varieties planted on 2 soil types - 1966.

Soil Type	Seed Infection	King Edward		Majestic		Redskin		Kerrs Pink	
		Average number of days to emergence	Blanking (per cent)	Average number of days to emergence	Blanking (per cent)	Average number of days to emergence	Blanking (per cent)	Average number of days to emergence	Blanking (per cent)
Med-loam	Severe, all eyes	66.2	31.0	69.7	43.0	64.7	21.0	55.4	2.1
	Trace or Free, no eyes	50.0	0	66.8	8.3	57.4	2.1	50.0	0
Sandy	Severe, all eyes	47.7	35.0	49.8	21.0	48.7	16.0	37.9	0
	Trace or Free, no eyes	35.8	2.1	37.1	0	39.6	0	35.3	2.1

Table 26. The effects on skin spot development in the resulting crop of different levels of skin spot infection on seed tubers of 4 varieties planted on 2 soil types - 1966.

Soil Type	Seed Infection	King Edward		Majestic		Redskin		Kerrs Pink	
		SII	EII	SII	EII	SII	EII	SII	EII
Med-loam	Severe, all eyes	8.6	40.0	10.7	34.1	13.4	43.3	11.1	46.5
	Trace or Free, no eyes	4.7	22.5	6.0	25.2	4.7	13.9	7.7	28.1
Sandy	Severe, all eyes	4.6	24.2	1.9	5.9	4.2	15.6	3.0	11.6
	Trace or Free, no eyes	1.7	5.2	2.2	6.0	0.8	1.2	1.7	4.2

Discussion

The results show that the rates of plant emergence where seed tubers of different skin spot infection categories are planted, is increasingly delayed as the level of infection on the seed tuber increases but that the effects of the disease may be modified by soil types and variety. Thus, in general, emergence was more rapid in a light sandy soil than in a medium-loam soil and in 2 of the 3 years of experimentation emergence rates for King Edward seed of different levels of infection planted in a sandy soil were similar. On the other hand more heavily infected seed invariably showed a slower rate of emergence than slightly infected seed or seed free from infection when planted in a medium-loam soil. Of the varieties examined, namely King Edward, Kerrs Pink, Majestic and Redskin, Kerrs Pink was found to show the most rapid rate of emergence and the effects of high levels of skin spot infection in delaying emergence were less on this variety than on the others.

Blanking occurred to a significant extent only where seed with all eyes infected was planted. This incidence of blanking, however, tended to be lower on the light sandy soil than on the medium-loam soil, though it was still very substantial. Kerrs Pink showed an insignificant amount of blanking in 1966 compared with the other varieties, but the level of 27 per cent for all eyes infected seed in the medium-loam soil in 1965, though considerably less than in the

corresponding King Edward seed, does show that fairly extensive blanking could occur with this variety. However, results from field studies on rate of emergence and blanking, coupled with those from small scale studies demonstrating how Oospora pustulans infected and physically damaged eyes in Kerrs Pink tubers could still produce healthy sprouts, show that a vigorous sprouting variety, highly susceptible to skin spot, can overcome to a large extent the damaging effects of skin spot infection of seed tubers on emergence.

Yield estimates which were only carried out in one year indicated that where there was a high level of blanking as in severe surface, all eyes infected planted seed, then there was a considerably reduction in yield.

For all varieties on both soil types the results of this experimental work demonstrate clearly that, in general, the higher the level of infection on the planted seed, then the higher incidence of skin spot there will be on the resulting crop, although anomalous results with severely infected King Edward seed were noted and are difficult to explain. No differences were obvious in overall skin spot development in the crops from the different varieties, but in the 1966 experiment carry-over of disease from badly infected seed appeared to be greater in Redskin than in other varieties.

There is now considerable evidence that Oospora pustulans can persist in soils, the presence of the fungus in soils for periods up to 10 years after potato crops had been grown in them having been demonstrated (Hirst and Salt, 1956; Salt,

1958; Nagdy, 1962). It is not possible, therefore, without some knowledge of the level of soil inoculum to conclude that the higher skin spot infection found on tubers from a medium-loam soil compared with that found on tubers from a light sandy soil, where the level of infection on the seed planted was the same was entirely an environmental effect. The plots used in 1965 and 1966 had all carried potatoes in recent years and no estimate of the soil infectivity was possible, but in the 1964 experiment the medium-loam plot had never grown potatoes while the light sandy plot had been contaminated regularly with soil dumped from a potato shed. Since less skin spot developed on the crops from the light sandy soil where there was possible fungal contamination than on the crops on the medium-loam soil where no contamination was likely, then this can be considered as evidence that a medium-loam soil will favour skin spot infection more than a light sandy soil. Future work on this problem will require some estimate of the soil infectivity, possibly using potato seedlings as indicator plants in the method developed by Hirst, Salt and Hide (1963).

In conclusion it can be said that by growing a crop on light sandy soil compared to a medium-loam soil, the adverse effect of skin spot infection on emergence will be reduced to some extent and there is some evidence that disease transmission to the resulting crop from the planted seed will be less.

SECTION D

Carbohydrate nutrition of *Oospora pustulans* and its relationship to carbohydrate changes in potato tubers during storage.

Introduction

Owen (1919) reported that *Oospora pustulans* would grow better on a cooked vegetable medium than on an uncooked vegetable medium, while Kharkova (1961b) observed that the addition of sugar to a medium will enhance the growth of *Oospora pustulans*, and suggested that high sugar contents in tubers would favour high skin spot infection. It has also been reported that various biochemical changes occur within tubers infected by *Oospora pustulans* including the breakdown of starch to sugar (Gomolyako, 1959; Vartapetyan, 1962). In an attempt to gain more information about the carbohydrate nutrition of *Oospora pustulans*, studies were carried out on the growth of the fungus in various carbohydrate media in relation to nutrient uptake from the media and carbohydrate changes taking place in the media. In addition an examination was made of the sugar contents of varieties of different levels of susceptibility to skin spot stored under conditions known to be either favourable or unfavourable for skin spot infection. From these findings it was hoped that some connection could be established between the carbohydrate nutritional requirements of the fungal pathogen and the carbohydrate status of the host

when subjected to conditions which favoured skin spot infection.

The studies may be divided into 2 parts:

1. Growth of Oospora pustulans on various carbohydrate media.
2. Carbohydrate changes during storage in tubers of different potato varieties, lifted mature and immature, and subjected to different storage temperatures.

EXPERIMENTAL WORK

D.1. Growth of Oospora pustulans on various carbohydrate media.

1. Materials and Methods

A series of 3 investigations were carried out as follows:

1. A comparative study of the growth rates of Oospora pustulans on starch and glucose media.
2. A comparative study of the growth rates of Oospora pustulans on various carbohydrate media.
3. A comparison of the growth of Oospora pustulans on various carbohydrate media after 7 weeks incubation.

The growth rates of the fungus on the different media were compared by measuring the dry weight of mycelium at intervals during incubation. At these intervals analyses were also made of the media to determine any pH changes, the rate of utilisation of the media and the production of free reducing sugar in the non-reducing sugar media.

Media were prepared (30 g. carbohydrate in 1 l. of Czapek's acid mineral solution) and added to test tubes 6 ins. x $\frac{3}{4}$ in. at the rate of 9 ml. per tube. Following sterilisation a series of tubes for each medium was inoculated under aseptic conditions with a spore suspension of Oospora pustulans using 1 ml. of spore suspension per tube. The remaining tubes in each medium were uninoculated and used as controls. The tubes were randomly arranged in a wire basket and incubated at about 10°C.

The original media were analysed for the quantity of reducing sugar by the Hanes' Ferricyanide Micromethod and pH measurements also taken. At intervals during incubation samples of inoculated and control tubes for each culture medium were randomly selected and examined. The fungal mycelium in each inoculated tube was filtered off and placed with the filter pad in an oven at 90°C for 24 hours. After cooling the mycelium was removed from the filter pad and weighed to give its dry weight. The filtrate was made up to 10 mls. with distilled water, the pH measured and the reducing sugar content (expressed as mg./0.1 ml. of medium) estimated as previously described. In addition, for the non-reducing sugar media of sucrose and starch, the total sugar content of the media was analysed by acidic hydrolysis to break down the molecules into reducing sugar units, the quantities of which were then estimated as above. The same tests were carried out on the control tubes for each medium.

Details of the procedure in the 3 investigations are given below.

Investigation	Media analysed	No. of tubes analysed at each sampling time	Conc. of spore suspension	Time of sampling
1	Glucose Starch (not analysed form total carbohydrate)	3 inoculated + 1 control	0.97×16^6 spores/ml.	Weekly after 2 weeks until 6 weeks after inoculation

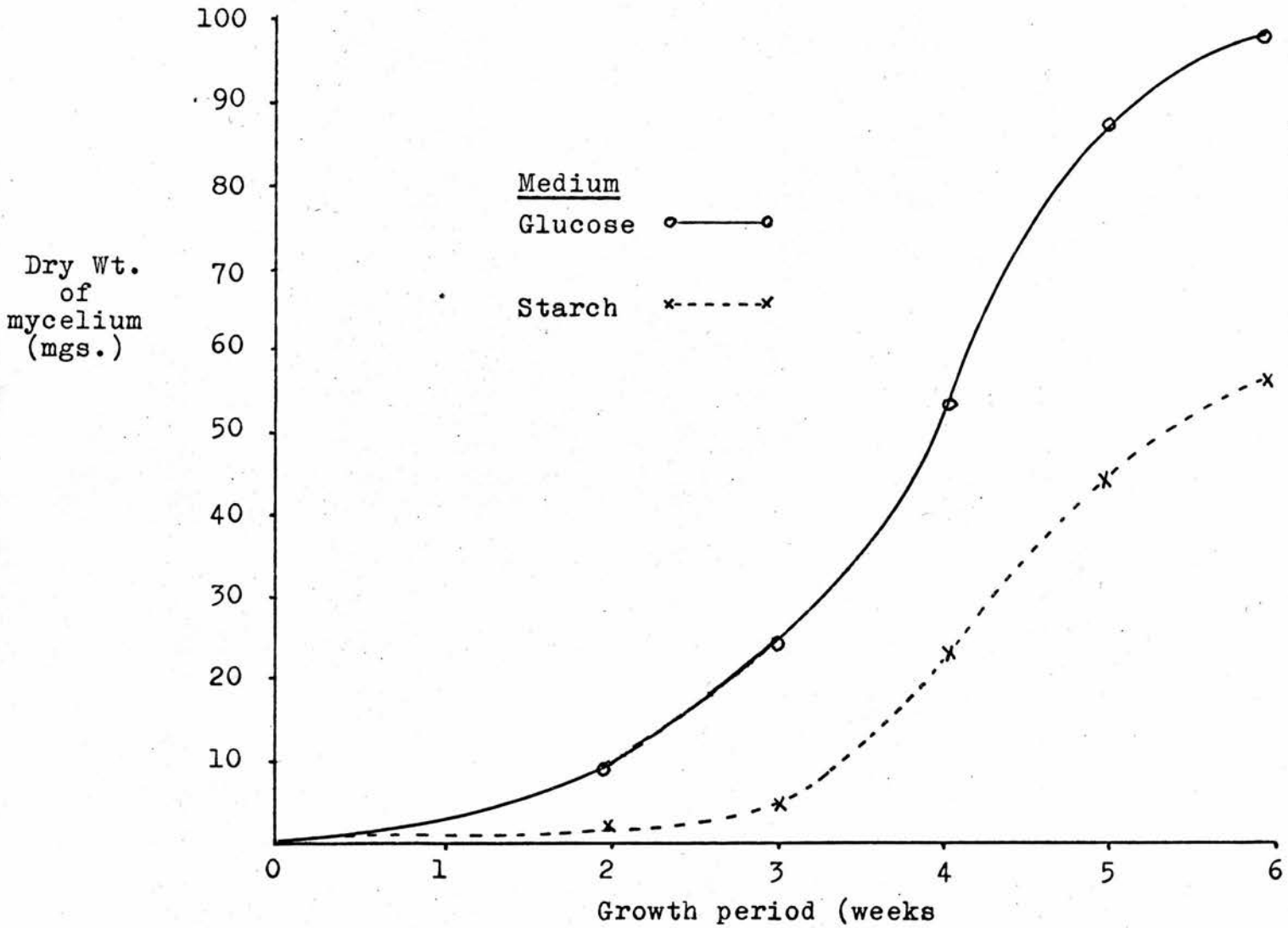
Investigation	Media analysed	No. of tubes analysed at each sampling time	Conc. of spore suspension	Time of sampling
2	Glucose Starch Sucrose Maltose	2 inoculated + 1 control	0.82×10^6 spores/ml.	Weekly up to 7 weeks after inoculation
3	Glucose Starch Sucrose Maltose Fructose	5 inoculated + 3 control	0.50×10^6 spores/ml.	Only after 7 weeks from inoculation

2. Results

2.1. A comparison of growth of Oospora pustulans on starch and glucose media

The results of the mycelial dry weight determinations at different stages after inoculation (Table 27, Fig. 11) indicate that the fungal growth was faster on a glucose medium than on a starch medium. For both growth curves there was an initial lag phase followed by a period of rapid growth and then a declining rate of growth. This latter phase was associated in the glucose medium with 93.2% of the glucose having been utilized by the 6th week (Table 27). There were indications that by the 5th and 6th weeks a slowing down in the growth of the fungus in the starch medium occurred, but no figures are available to relate this to changes in the medium. In the starch medium the initial lag phase was longer than in the glucose medium and the phase of rapid growth may be associated

Figure 11. Growth rate of Oospora pustulans in different carbohydrate media as measured by dry weight of mycelium.



with the appearance of free reducing sugar (Table 27). The figures for pH of the original media were about 5.0, but once growth was established these rose to about 7.5.

Table 27. Changes in dry weight of fungal mycelium and of the reducing sugar content of the media measured at weekly intervals during the incubation of Oospora pustulans in glucose and starch media.

Period of incubation (weeks)	Glucose medium			Starch medium	
	Dry wt. of mycelium (mgs)	Reducing sugar present (mg/0.1 ml of medium)	Medium utilised (per cent)	Dry wt. of mycelium (mgs)	Reducing sugar present (mg/0.1 ml of medium)
2	8	2.18	31.9	1	0
3	23	1.86	41.8	4	0.16
4	52	1.31	59.2	23	0.43
5	87	0.44	86.3	44	0.60
6	95	0.22	93.2	56	0.85

2.2. A comparison of growth of Oospora pustulans on various carbohydrate media

From these results (Table 28 and Figs. 12, 13) it can be seen that the fungus showed a greater increase in dry weight in the 3 sugar media than in the starch medium, the response to the 3 sugars being similar. The results of assessments of the percentage of the media which had been used up at the various

stages after inoculation suggest that the rate of utilization of the media followed the rate of growth for each medium more especially when growth was established. In the first 3 weeks, i.e. during the initial lag phase, the values for the percentage of each medium utilised were erratic, but they were still low compared to those found when growth was rapid. It is interesting to note that by the 7th week of incubation the sugar media had been almost completely utilised, but there was still about 45 per cent of the starch medium left.

Table 28 also shows the levels of free reducing sugar, expressed as a percentage of the total carbohydrate content of the original medium, produced in the sucrose and starch media during incubation.

In the sucrose medium reducing sugars were produced by the second week and increased until by the fifth week almost all the sucrose remaining had been hydrolysed, since only 49.5% of the carbohydrate remained and the amount of reducing sugar present in this was 47.3% of the original medium. In the starch medium a certain level of reducing sugar was found from the second week onwards. This level at the second week was not such a high percentage of the original medium as it was at the corresponding stage in the sucrose medium. The reducing sugar rose in the starch medium until by the fifth week it accounted for about 50% of the unused medium, 63% of the original medium remaining and 37% of the original medium being in the form of reducing sugar. This production of reducing sugar in the sucrose and starch media might be attributed to

Table 28. Changes in dry weight (mgs.) of fungal mycelium, of the quantity of medium utilised (per cent of original medium) and in the level of free reducing sugar produced in non-reducing sugar media (per cent of original medium) measured at weekly intervals during the incubation of Oospora pustulans in different carbohydrate media.

Period of incubation (weeks)	Glucose medium		Maltose medium		Sucrose medium			Starch medium		
	Dry weight of mycelium	Medium utilised	Dry weight of mycelium	Medium utilised	Dry weight of mycelium	Medium utilised	Free reducing sugar present	Dry weight of mycelium	Medium utilised	Free reducing sugar present
1	0	5	0	0	0	24	0	0	16	0
2	1	13	1	2	1	7	51	0	14	11
3	9	15	4	23	12	20	73	7	11	22
4	27	31	28	29	24	24	57	17	22	23
5	57	56	48	58	45	52	55	35	37	32
6	56	67	59	65	58	65	42	43	49	31
7	87	89	72	88	88	83	16	52	55	27

Figure 12. Growth rate of Oospora pustulans in different carbohydrate media as measured by dry weight of mycelium.

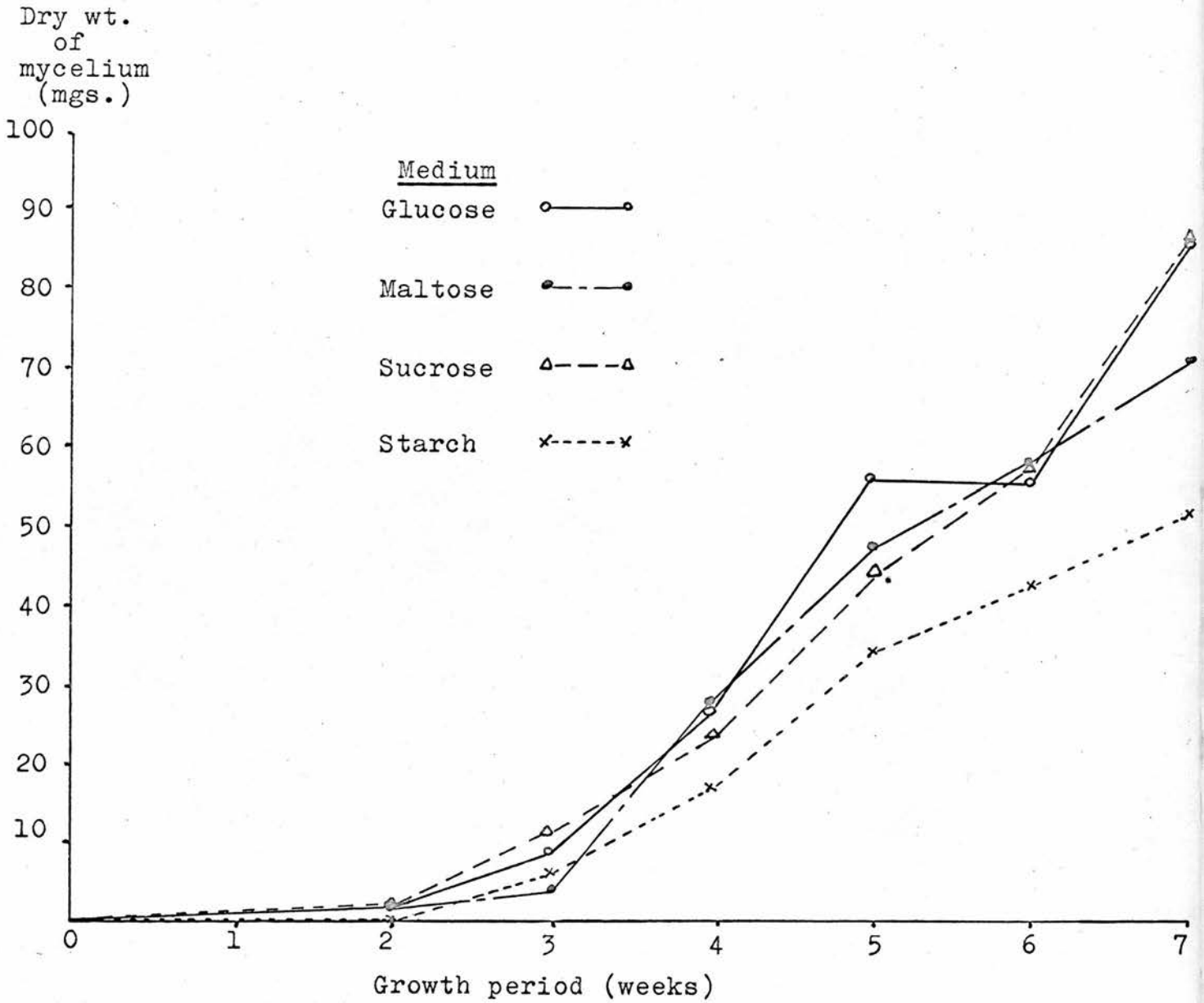
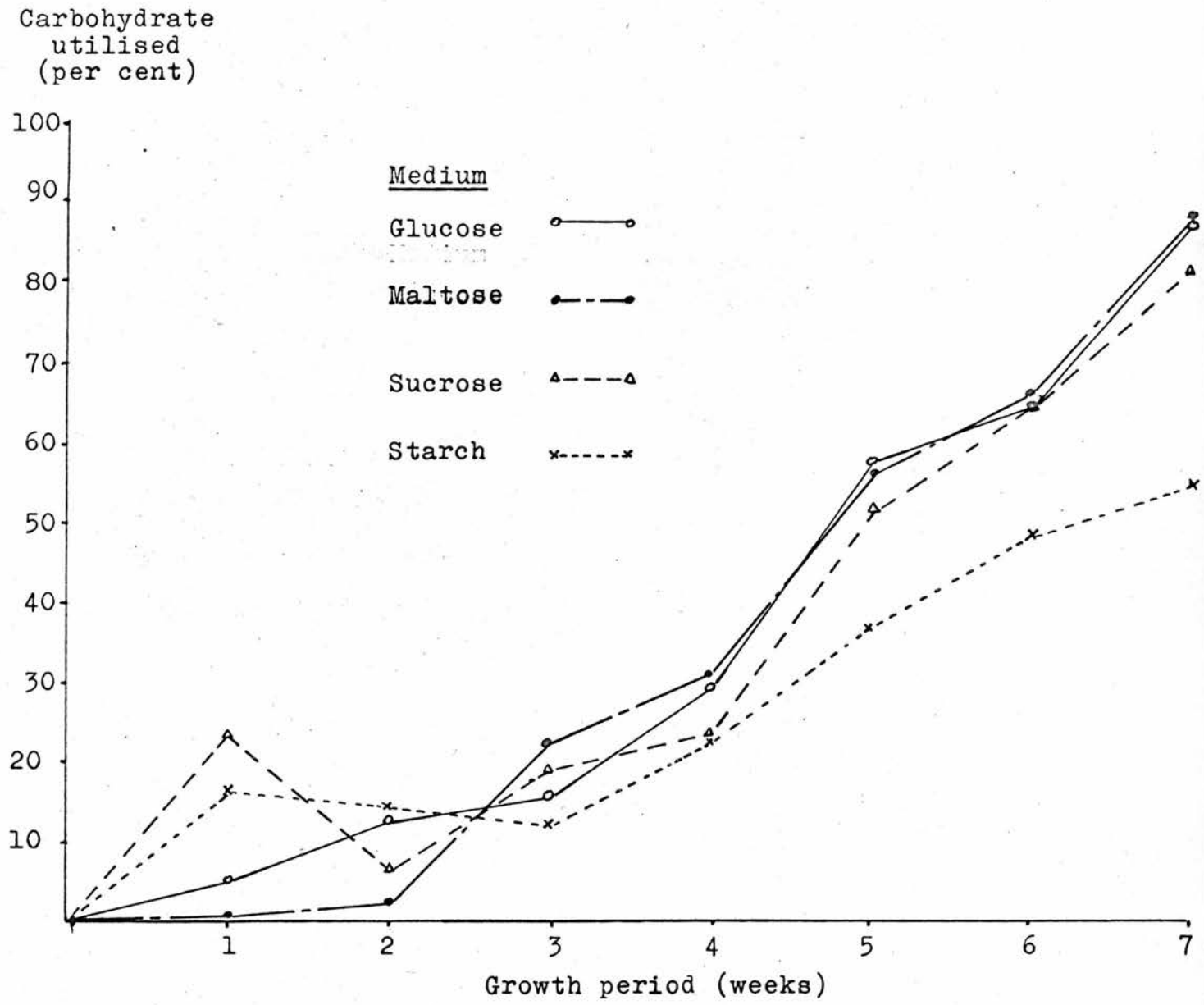


Figure 13. Quantities of the carbohydrate content of different media utilised (per cent of original medium) at weekly stages during the incubation of Oospora pustulans.



autolytic breakdown of the molecules, but analysis of control tubes at each sampling stage showed only a negligible production of reducing sugars as the age of the medium increased. It might also be argued that the breakdown of these molecules related to changes in pH which increased from about 5.0 in the original medium to about 7.5 when growth was established. However, hydrolysis of starch and sucrose by chemical means is associated with increasing acidity rather than a rise in pH and hence it is unlikely that the pH changes in the media, which were about neutral when reducing sugars were most readily produced, accounted for the breakdown of starch and sucrose. It is also unlikely that the reducing sugars are produced by autolytic breakdown of the fungus since 50.7% of the original sucrose medium was in the form of reducing sugars by the second week after inoculation, yet growth of the fungus was still negligible and moreover the period when reducing sugars were most readily produced coincided with a period of active growth of the fungus in both media. The most probable explanation is that the fungus itself brings about the breakdown of these molecules by the production of hydrolytic exoenzymes and utilises carbohydrates in the form of monosaccharides. This theory is in keeping with the results which show that the production of reducing sugar was associated with the period of most active growth in the sucrose and starch media, indicating that it was the availability of these basic sugar units which was allowing growth to increase. Moreover, there is proof that the fungus

does use the hydrolysed sugar units, since with the sucrose medium in the last 2 weeks of incubation almost 100% of the remaining medium was reducing sugar and growth was extremely active.

2.3. A comparison of the growth of Oospora pustulans on various carbohydrate media after 7 weeks of incubation

Growth of the mycelium as determined by its dry weight after 7 weeks (Table 29) was less on the starch medium than on all the sugar media with the exception of maltose on which growth was also of a relatively low order while glucose appeared to give the largest dry weight increase. The amount of medium utilised agreed reasonably well with the amount of growth. The levels of free reducing sugars in the starch and sucrose media were similar to those found in the previous experiments, but apart from the glucose medium the general level of growth in the media was not as high as in the previous experiment. This may be due to the smaller spore concentration in the inoculum for this experiment.

Table 29. Dry weight of fungal mycelium (mgs.), the quantity of medium utilised (percentage of original medium) and the level of free reducing sugar produced in non-reducing sugar media (percentage of original medium) after 7 weeks incubation of Oospora pustulans in different carbohydrate media.

Medium	Glucose	Fructose	Maltose	Sucrose	Starch
Dry weight of mycelium (mg)	98	55	45	69	44
Medium utilised (percent)	95	77	69	76	45
Free reducing sugar present (percent)	-	-	-	22	37

D.2. Carbohydrate changes during storage in tubers of different potato varieties, lifted mature and immature, and subjected to different storage temperatures.

1. Materials and Methods

In this experiment analyses were made periodically during storage of the sugar content of tubers of different varieties with varying degrees of susceptibility to skin spot infection (Boyd and Lennard, 1961a; Nagdy and Boyd, 1965), lifted in immature and mature states and subjected to different storage temperatures, both factors being known to have some effect on infection.

Tubers of 5 varieties, Kerrs Pink, King Edward and Arran Banner (highly susceptible to skin spot) and Arran Consul and Golden Wonder (highly resistant to skin spot) were lifted on 2 dates, 10th September 1965 and 4th October 1965. On each date tubers were randomly selected from the drills and for each variety 5 tubers of about seed size were placed in each of 12 small cardboard boxes. Four boxes of each variety were then allocated for storage under 3 temperature regimes:

(a) About 15°C;

(b) About 5°C;

(c) Fluctuating temperature in an insulated shed (0°C - 18°C).

During storage the boxes were kept moist by periodic spraying with water. At lifting and at monthly intervals until 4 months after lifting one box for each variety, mature

and immature, from the different temperature treatments, was removed and the sugar contents determined as described in General Materials and Methods.

2. Results

The figures on Table 30 show the total sugar content of the tubers as mg./100 g. of fresh weight at monthly intervals after lifting in relation to variety, time of lifting and storage temperature. Figure 14 and Appendix XXXII show the mean values of the sugar contents in the 3 temperature regimes for each variety, mature and immature, and Figure 15 and Appendix XXXIII show the mean values of the sugar contents in the 5 varieties for each temperature regime at monthly intervals after lifting and for mature and immature tubers.

While there were slight differences in sugar contents between the different varieties at the beginning of storage (Fig. 14) these were not marked and for all varieties the sugar contents of the tubers on both lifting dates were relatively low with the earlier lifted samples having slightly less than the later lifted samples. No consistent pattern of difference in production of sugars could be seen between the varieties during the storage period for either the immature or mature lifted tubers.

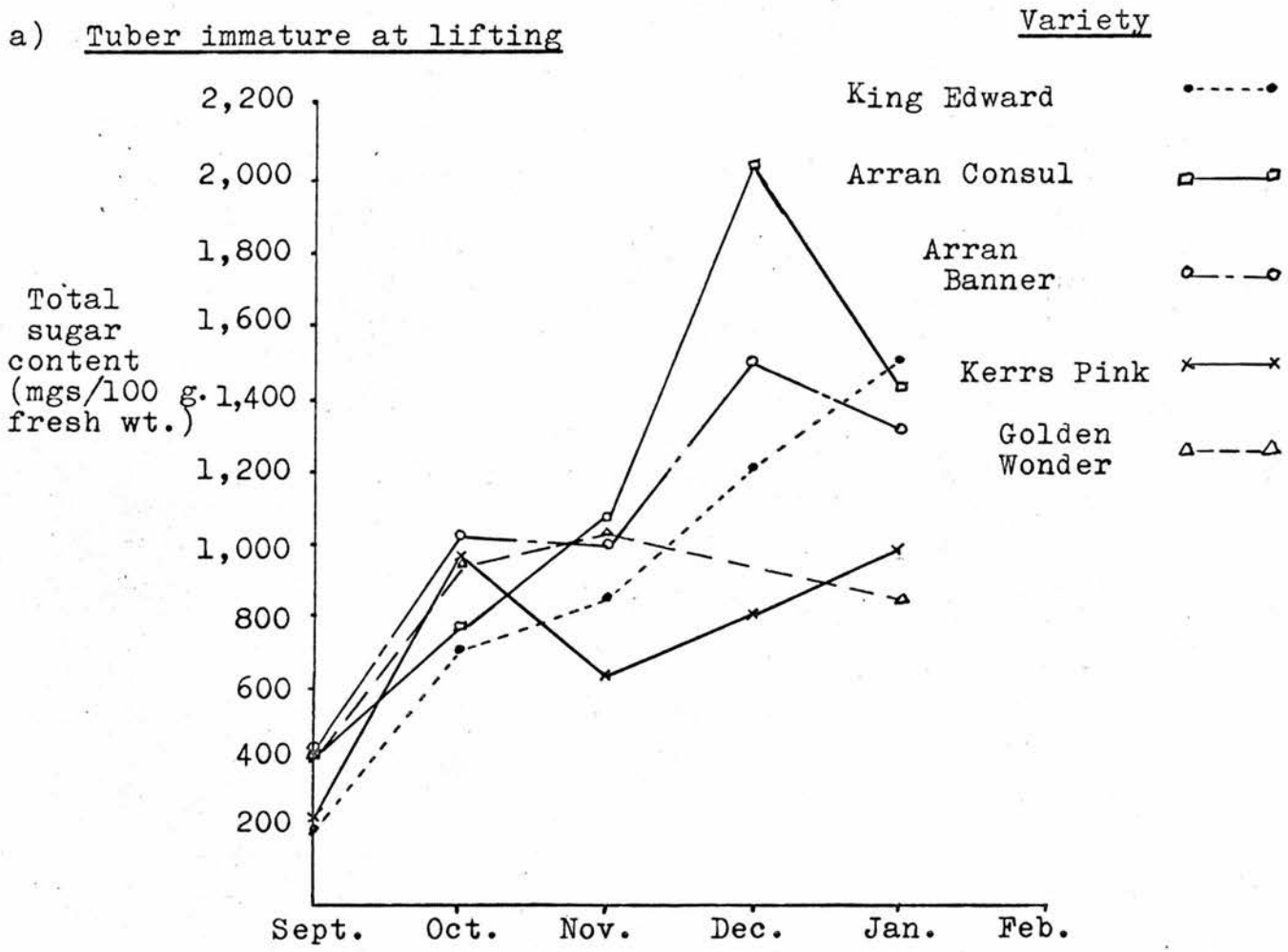
Where tubers were stored at 15°C (Fig. 15) there were no marked changes in levels of sugar content from the time of lifting onwards. On the other hand in tubers stored at 5°C there was a considerable increase in sugar content which was

Table 30. Total sugar content (mg./100 g. fresh weight) of tubers of different varieties lifted mature and immature and stored under different temperature conditions, measured at monthly intervals.

Variety	Storage Temperature	Total sugar content at monthly intervals											
		Sept.		Oct.		Nov.		Dec.		Jan.		Feb.	
		Imm.	Mat.	Imm.	Mat.	Imm.	Mat.	Imm.	Mat.	Imm.	Mat.	Imm.	Mat.
Kerrs Pink	15°C	248	-	392	530	136	296	190	260	146	236	-	332
	5°C	248	-	2370	530	1525	1140	1030	1480	2270	1330	-	750
	Fluctuating (0-18°C)	248	-	170	530	260	1160	1180	980	540	1860	-	755
King Edward	15°C	235	-	262	285	277	124	124	300	93	206	-	246
	5°C	235	-	1682	285	1790	524	2520	1350	2205	1280	-	1970
	Fluctuating (0-18°C)	235	-	210	285	470	918	930	830	2260	1060	-	854
Arran Banner	15°C	436	-	490	660	529	653	472	771	152	615	-	893
	5°C	436	-	2220	660	1610	1112	2770	1750	2280	835	-	1880
	Fluctuating (0-18°C)	436	-	675	660	860	2030	1280	1430	1510	2450	-	1402
Arran Consul	15°C	404	-	480	681	350	900	416	428	328	380	-	357
	5°C	404	-	1518	681	2120	2340	3210	2380	3130	3060	-	1480
	Fluctuating (0-18°C)	404	-	360	681	1080	2640	2520	2070	780	2500	-	1780
Golden Wonder	15°C	400	-	755	675	234	504	-	204	192	97	-	406
	5°C	400	-	1522	675	2360	1430	-	1820	1075	1225	-	2850
	Fluctuating (0-18°C)	400	-	675	675	490	1910	-	1050	1160	960	-	688

Figure 14. Mean value of the total sugar content of tubers in 3 temperature regimes of each of 5 varieties, lifted mature and immature, measured at monthly intervals during storage.

a) Tuber immature at lifting



b) Tubers mature at lifting

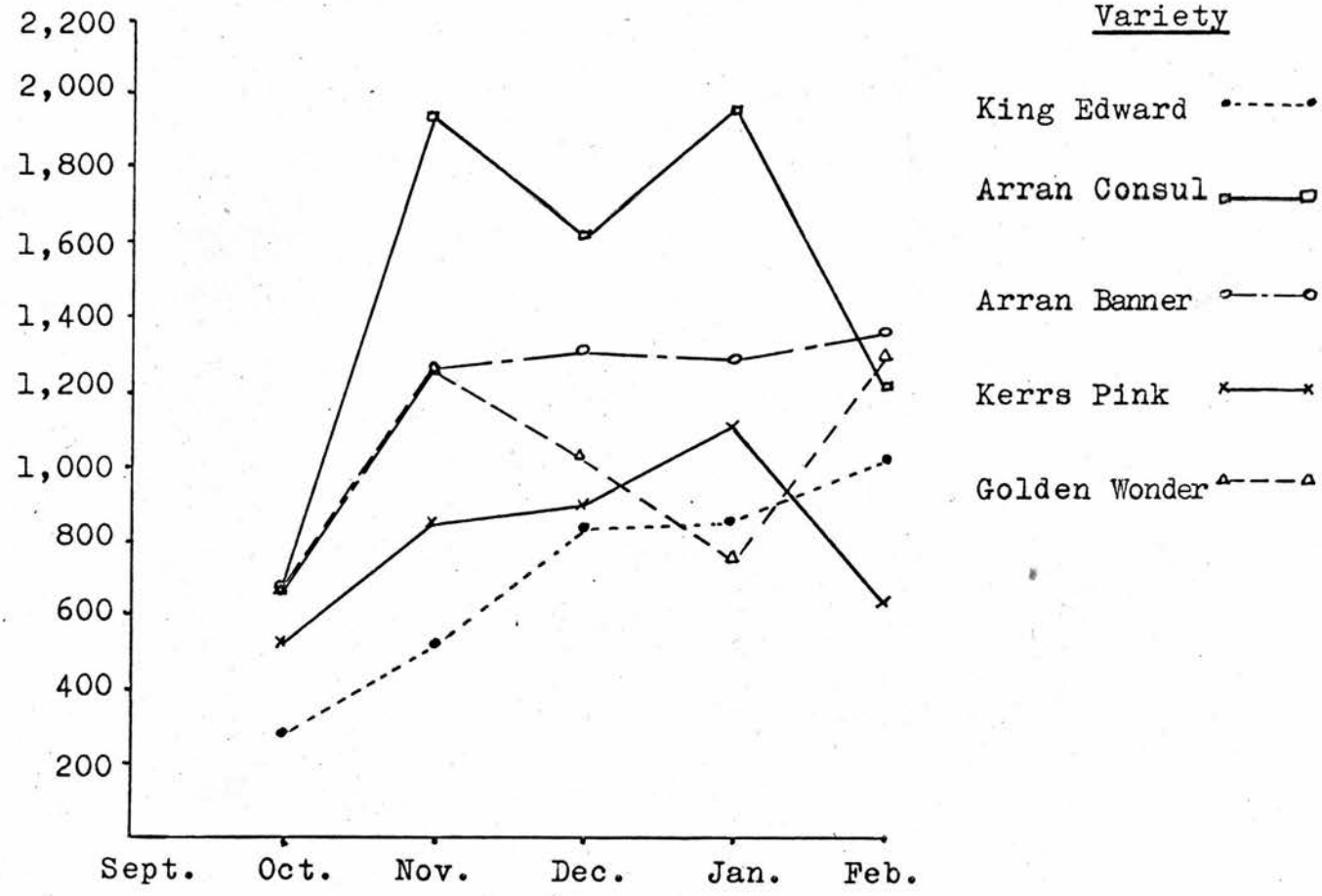
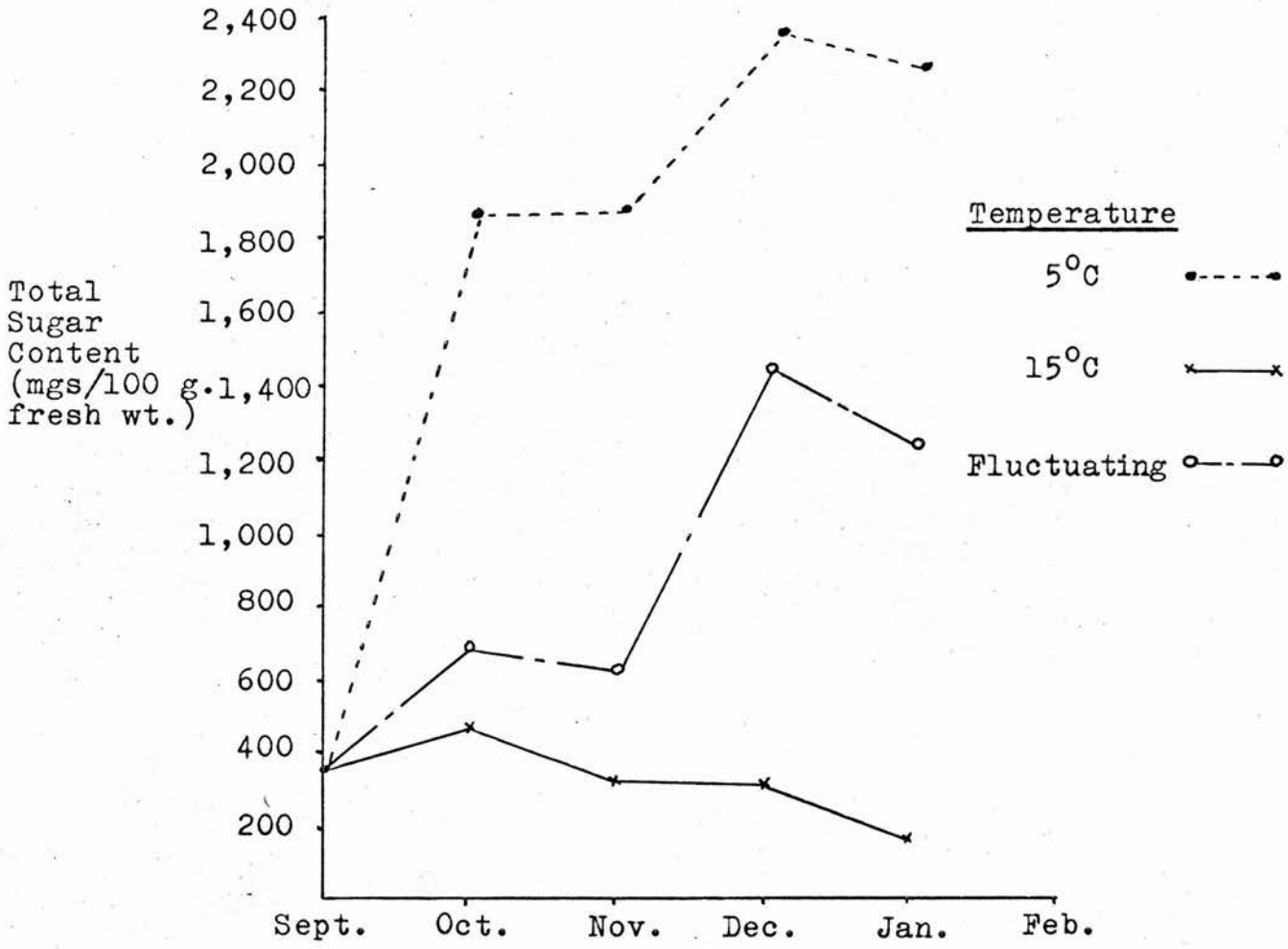
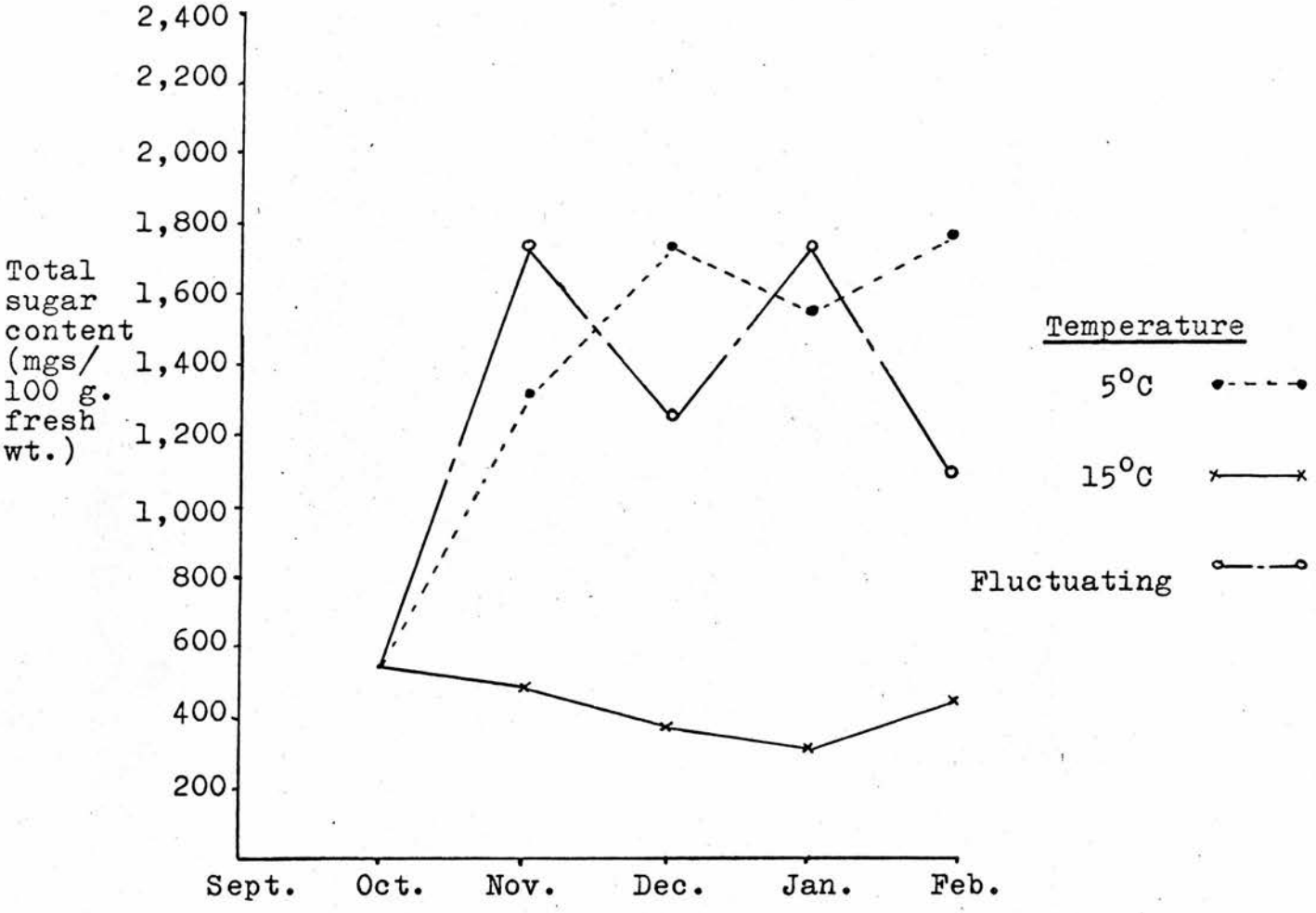


Figure 15. Mean value of the total sugar content of tubers in 5 varieties for each of 3 temperature regimes, after lifting immature and mature, measured at monthly intervals during storage.

a) Tubers immature at lifting



b) Tubers mature at lifting



evident in the analysis made after the first month of storage both for early- and late-lifted tubers. In the tubers kept in the insulated shed, subject to ambient air temperature the early- and late-lifted tubers showed a slightly different response in the early part of storage, although in both cases the sugar contents were eventually similar to those produced by continual storage at 5°C. With the late-lifted tubers the sugar content increased to a high level within the first month of storage as is shown from the results for November. With the early-lifted tubers, however, such a level of sugar production was not reached until 3 months of storage, i.e. in December. The delayed increase in sugar content in early-lifted tubers in storage at fluctuating temperatures may be attributed in part to higher temperatures in the first month of storage. However, the slower rise in sugar content in early-lifted tubers compared with that in late-lifted tubers when they were both stored under the same conditions would suggest a carry-over effect from lifting tubers immature although the reasons for this are not clear. The responses of the different varieties to the various temperature regimes are clearly seen to have been similar (Table 30).

Discussion

The evidence from the growth studies of Oospora pustulans on various carbohydrate media indicates that the fungus will grow at a faster rate on a sugar medium than on a starch medium. Between the different sugar media tested, glucose, sucrose, maltose, fructose, there were no marked differences in growth response, although growth on maltose tended to be unexpectedly low in one test. It is also clear that the fungus is capable of breaking down poly- and disaccharide molecules into monosaccharide units and that this breakdown is associated with the period of active growth of the fungus. The slower rate of utilisation of the starch medium and the resulting slower rate of growth compared with that on the other media may be attributed to the delay involved while glucose, units required for fungal growth, are produced by biological degradation of the starch molecules. The rapid production of monosaccharide units in the sucrose media is not surprising since sucrose molecules require hydrolysis of only one glycosidic link, a relatively simple biochemical process compared with the breakdown of complicated starch molecules. Thus no delay in utilisation of sucrose compared with the other media was experienced.

The experimental work in Section A shows that later dates of lifting and low temperature storage are found to favour skin spot development. The results from this investigation indicate that such conditions are associated with high sugar

contents in the tubers which might in turn favour growth of Oospora pustulans. However, the lack of any correlation between susceptibility of different varieties to skin spot and their sugar content would suggest that the content of sugar in the tuber is not a primary factor in determining susceptibility to the disease. This does not exclude the possibility of this factor coming into play in susceptible varieties, but to what extent it can influence fungal infection is not clear. The results of this work, however, are in keeping with the findings of Kharkova (1961b) that high sugar contents in tubers would favour high skin spot infection.

GENERAL DISCUSSION

From the studies of factors involved during lifting in relation to skin spot development, the results would suggest that satisfactory control of the disease may be obtained by boxing tubers at lifting time but only where lifting is carried out early, when ambient air temperatures are relatively high in the first few weeks of storage. At later dates of lifting only disinfection in an organo-mercury solution, coupled with boxing, gave any effective measure of control. This treatment was also effective when carried out after lifting and clamp storage but only up until a few weeks after lifting, coinciding with the maximum depth of penetration of the fungus into the tissues. Attempts to find satisfactory alternatives to organo-mercury solution showed that none of the fungicides tested proved as effective as mercury and could not be recommended in practice. While Boyd (1957) suggested that early haulm destruction may be beneficial in reducing skin spot development the present work would suggest that this treatment would not prove consistently effective in practice. These studies do agree, however, with the work of Greeves and Muskett (1939) where early lifting and clamp storage did not effectively reduce skin spot development and that of Boyd (1957) where early lifting and box storage did reduce the disease.

When tubers which had been boxed and disinfected in organo-mercury solution and boxed only at lifting were planted, they both showed better emergence than seed which had been

clamped until December or continuously throughout storage despite the fact that the boxed only tubers had significantly more skin spot infection than the disinfected tubers, though slightly less than the clamped tubers, but it was found that crops from the seed which had been disinfected developed less skin spot than those from seed which had been boxed only or clamped. Disinfecting with organo-mercury at planting also reduced disease development in the resulting crop though it did not appear to be as effective as disinfecting at lifting, thus agreeing with Greeves and Muskett (1939) and Edie (1964). Moreover it was apparent, possibly due to a phytotoxic effect on the sprouts, that there was a delay in emergence caused by disinfecting at planting, as was also found by Boyd and Lennard (1963).

During storage a progressive development of infection appears to occur under humid and cool conditions; however, as artificial inoculation after late lifting gave no increase in number of dead eyes as compared with natural infection from the field and only a small increase in the number of infection sites, any secondary spread of infection would appear to be superficial in nature and have no marked effect on eye damage. When sprouted Kerrs Pink tubers were artificially inoculated in mid-November, however, considerable eye damage was caused by the fungus, but in this case the buds were abnormally well developed and were likely to be more sensitive to fungal infection. Under conditions favourable to disease development there was a significant increase in infected dead eyes at the 12-16 week period

after lifting, this was considered to be caused by hitherto infected live eyes being killed. Theoretically, if tubers were sprouted just before this period many of the eyes might survive the infection. An experiment which examined such a treatment showed that while the rate of emergence was higher for the sprouted tubers than for the unsprouted, no differences in blanking or yield were evident. However, the disease level on the seed in the experiment was of a relatively low order and with a more severe infection some advantage from sprouting might have been evident.

Boyd and Lennard (1962) suggested that factors underlying skin spot development were rainfall during the lifting period, temperature during storage and the level of seed inoculum. Each of these factors have been considered in some detail in the work undertaken here.

Although Boyd and Lennard indicated that above average rainfall at lifting gave a high disease incidence, from studies on the effects of different watering rates to the soil prior to lifting on subsequent skin spot development the results suggested that moderate rather than high levels of soil moisture favoured skin spot infection. Further work is required to confirm these results especially in view of the evidence that a high humidity in storage which may be associated with wet soil conditions is an important factor contributing towards a high level of disease development.

The importance of storage temperature in relation to skin spot development has been shown in field and small scale

studies. The temperature levels over the lifting period and in the first few weeks of storage appear to be particularly important. The effect of a low temperature in encouraging infection may be associated with the defence mechanism of the host in delaying the production of a cork barrier to seal off the fungus or may also be due to more active growth of the pathogen, but probably both these factors are involved. The observation made in this work that growth of the fungus appeared to be more vigorous in infected tissue at 5°C than 15°C and those of previous workers where abnormally large lesions occurred in tubers and no cork barrier was formed (Ives, 1955; Boyd and Lennard, 1961b) would favour the theory that it is a combination of these factors.

The effect of level of seed inoculum on skin spot infection in the subsequent crop has been investigated by Boyd and Lennard (1961a) and Hirst et al. (1967) who indicated that a high level of inoculum on King Edward, Arran Pilot and Majestic seed was associated with a high infection of the resulting crop. This work has confirmed these findings and has also established that other varieties react in a similar manner regarding disease transmission. There is also tentative evidence to suggest that infection is less favoured on a light sandy soil than it is on a medium-loam soil. Boyd and Lennard (1961b) also examined the field effect of emergence in relation to the levels of infection on the seed, finding that emergence was delayed as the level of seed infection increased. These findings were confirmed in the results of

this further work which, however, also indicated that the adverse effects of infection on emergence are less in vigorous sprouting varieties or for seed planted in lighter soils.

The investigation of the carbohydrate nutrition of Oospora pustulans showed that tubers subjected to conditions of later dates of lifting and low temperature storage, which were known to favour skin spot development, also had high sugar contents thus favouring growth of the fungus. However, high sugar contents are developed by non-susceptible varieties in the conditions described above. This would suggest that sugar content is not a primary factor in skin spot development in susceptible varieties although it may be an important factor in promoting more active growth of the fungus when conditions favour disease development.

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Appendix I

Skin spot surface and eye infection indices in relation to time of haulm destruction, time of lifting and storage treatment - 1965.

Date of Haulm Destruction	Date of Lifting	Storage Treatment							
		Boxed and disinfected		Boxed and washed		Boxed alone		Clamped	
		SII	EII	SII	EII	SII	EII	SII	EII
23 Aug.	23 Aug.	0.5	6.0	1.3	17.4	1.5	20.3	-	-
	6 Sept.	0.8	11.6	1.9	26.8	2.5	33.8	5.8	48.6
	23 Sept.	1.0	19.6	4.4	48.5	3.0	37.6	10.5	51.0
	6 Oct.	0.7	10.9	2.9	40.8	3.5	40.9	7.6	48.1
	19 Oct.	1.0	25.0	4.7	51.1	4.3	49.1	7.1	44.0
6 Sept.	6 Sept.	0.6	7.9	2.1	27.5	2.1	31.3	-	-
	23 Sept.	0.8	18.8	4.0	37.8	3.0	35.7	5.2	38.2
	6 Oct.	0.7	13.6	3.9	40.0	3.8	40.5	6.1	41.7
	19 Oct.	1.1	17.1	6.5	46.5	3.7	51.3	8.0	50.5
23 Sept.	23 Sept.	0.7	12.2	3.2	40.9	3.0	41.5	-	-
	6 Oct.	0.4	8.4	2.7	32.8	4.0	37.1	5.8	40.6
	19 Oct.	0.5	12.3	4.6	43.5	3.4	42.3	4.0	34.1

Appendix II

Skin spot surface and eye infection indices in relation to time of haulm destruction, time of lifting and storage treatment - 1966.

Date of Haulm Destruction	Date of Lifting	Storage Treatment					
		Boxed and disinfected		Boxed alone		Clamped	
		SII	EII	SII	EII	SII	EII
23 Aug.	23 Aug.	0.4	4.4	1.0	10.8	-	-
	6 Sept.	0.4	4.4	1.7	22.8	3.9	26.4
	17 Oct.	1.1	17.4	4.3	37.9	-	-
6 Sept.	6 Sept.	0.3	5.6	1.3	19.2	-	-
	20 Sept.	0.2	1.7	2.3	28.3	4.6	30.8
	17 Oct.	1.1	15.3	5.9	39.6	-	-
20 Sept.	20 Sept.	0.2	6.3	2.1	24.5	-	-
	4 Oct.	0.6	12.2	2.6	24.8	6.0	32.1
	17 Oct.	0.7	10.8	4.9	35.6	-	-
Natural Senescence	4 Oct.	0.4	7.5	3.0	30.6	-	-
	17 Oct.	1.4	17.4	6.0	38.5	6.3	31.2

Appendix III

Skin spot surface and eye infection indices in relation to time of haulm destruction and storage treatment of tubers lifted in mid-October 1965 and 1966.

Year	Infection Index	Storage Treatments and Dates of Haulm Destruction															
		Boxed and disinfected				Boxed and washed				Boxed alone				Clamped			
		23 Aug.	6 Sept.	20 or 23 Sept.	Natural Senescence	23 Aug.	6 Sept.	20 or 23 Sept.	Natural Senescence	23 Aug.	6 Sept.	20 or 23 Sept.	Natural Senescence	23 Aug.	6 Sept.	20 or 23 Sept.	Natural Senescence
1965	SII	1.0	1.1	0.5	-	4.7	6.5	4.6	-	4.3	3.7	3.9	-	7.1	8.0	4.0	-
	EII	25.0	17.1	12.3	-	51.1	46.5	43.5	-	49.1	51.3	42.3	-	44.0	50.5	34.1	-
1966	SII	1.1	1.1	0.7	1.4	-	-	-	-	4.3	5.9	4.9	6.0	-	-	-	6.3
	EII	17.4	15.3	10.8	17.4	-	-	-	-	37.9	39.6	35.6	38.5	-	-	-	31.2

Appendix IV

Skin spot surface and eye infection indices, averaged for different times of haulm destruction, in relation to time of lifting and storage treatment of tubers.

Year	Infection Index	Storage Treatments and Dates of Lifting																			
		Boxed and disinfected					Boxed and washed					Boxed alone					Clamped				
		23 Aug.	6 Sept.	20 or 23 Sept.	4 or 6 Sept.	17 or 19 Oct.	23 Aug.	6 Sept.	23 Sept.	6 Oct.	19 Oct.	23 Aug.	6 Sept.	20 or 23 Sept.	4 or 6 Oct.	17 or 19 Oct.	23 Aug.	6 Sept.	20 or 23 Sept.	4 or 6 Oct.	17 or 19 Oct.
1965	SII	0.5	0.6	0.8	0.6	0.9	1.3	2.0	3.9	3.2	5.3	1.5	2.3	3.0	3.8	4.0	-	5.8	7.8	6.5	6
	EII	6.0	9.7	16.9	11.9	18.1	17.4	27.1	42.4	37.8	47.0	20.3	32.5	38.9	39.5	47.6	-	48.5	44.6	43.5	4
1966	SII	0.4	0.4	0.2	0.5	1.1	-	-	-	-	-	1.0	1.5	2.2	2.8	5.3	-	3.9	4.6	6.0	
	EII	4.4	5.0	4.0	9.8	15.2	-	-	-	-	-	10.8	21.0	26.9	23.7	37.9	-	26.4	30.8	32.1	3

Appendix V

The effects of time of haulm destruction, time of lifting, and storage treatment on skin spot development - 1965-66.

Comparison of treatment means using Students 't' test

Comparison number	Lifting date	Haulm destruction date	Storage treatment	Surface or eye infection	Number in sample	Mean	Arcsin transformation mean	Difference of means	S.E. of difference	t	Probability at t significant
a	17 Oct. 1966	23 Aug.	Boxed	Surface	3	4.34	12.01	2.05	1.58	1.30	-
		Natural Senescence			3	6.01	14.06				
b	17 Oct. 1966	Natural Senescence	Disinfected	Surface	3	1.42	6.63	4.16	1.61	2.56	-
	20 Sept. 1966	6 Sept.			3	0.19	2.47				
c	19 Oct. 1966	23 Aug.	Washed	Eye	2	48.5	-	10.7	7.20	1.49	-
		6 Sept.			2	37.8					
d	17 Oct. 1966	20 Sept.	Boxed	Eye	3	37.6	-	4.0	7.49	0.53	-
		4 Oct.			3	35.6					
e	17 Oct. 1966	4 Oct.	Disinfected	Eye	3	10.8	-	6.6	4.82	1.37	-
		Natural Senescence			3	17.4					

Appendix V (contd.)

Comparison number	Lifting date	Haulm destruction date	Storage treatment	Surface or eye infection	Number in sample	Mean	Arcsin transformation mean	Difference of means	S.E. of difference	t	Probability at t significant
f	23 Aug. 1965	23 Aug. 1965	Boxed	Eye	2	20.3	-	13.8	5.94	2.32	-
			Disinfected		2	6.5					
g	23 Sept. 1965	23 Aug. 1965	Boxed	Eye	2	39.6	-	20.0	4.58	4.36	0.05
			Disinfected		2	19.6					
h	6 Sept. 1965	23 Aug. 1965	Boxed	Surface	2	2.5	9.08	3.94	0.63	6.25	0.05
			Disinfected		2	0.8	5.14				
i	23 Aug. 1965	23 Aug. 1965	Boxed	Surface	2	1.5	6.88	2.61	1.47	1.77	-
			Disinfected		2	0.5	4.27				
j	23 Sept. 1965	23 Aug. 1965	Boxed	Surface	2	3.0	9.86	4.37	0.69	6.33	0.05
			Disinfected		2	1.0	5.59				
k	17 Oct. 1966	23 Aug. 1966	Boxed	Surface	3	4.34	12.01	5.95	0.82	7.24	0.01
			Disinfected		3	1.14	6.06				

Appendix VI

The effects of time of lifting on the skin spot development in tubers stored in clamps after lifting - 1966.

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Treatment	3	18.7045	6.2348	3.70	4.07	7.59	15.83
Error	8	13.4791	1.6848				
Total	11	32.1836					

Appendix VII

The effects of time of lifting on the skin spot development in tubers boxed only at lifting - 1965, 1966.

Analysis of Variance of surface infection 1965 (arcsin transformation)

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Treatment	4	47.4381	11.859	14.017***	2.90	4.50	7.26
Error	19	16.0795	0.084				
Total	23	63.5176					

Analysis of Variance of surface infection 1966 (arcsin transformation)

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Treatment	4	1594.93	398.73	5.84	2.71	4.07	6.25
Error	28	1910.85	68.24				
Total	32	3505.78					

Analysis of Variance of eye infection 1965

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Treatment	4	1379.57	344.89	12.56***	2.90	4.50	7.26
Error	19	521.56	17.45				
Total	23	1901.13					

Analysis of Variance of eye infection 1966

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Treatment	4	2368.50	592.12	16.48***	2.71	4.07	6.25
Error	28	1006.00	35.93				
Total	32	3374.50					

Appendix VIII

The effects of time of lifting on the skin spot development in tubers disinfected and boxed at lifting - 1965, 1966.

Analysis of Variance of eye infection 1965

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Treatment	4	390.67	97.66	4.24*	2.90	4.50	7.26
Error	19	440.83	23.20				
Total	23	831.50					

Analysis of Variance of eye infection 1966

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Treatment	4	774.93	193.77	10.43***	2.71	4.07	6.25
Error	28	519.91	18.56				
Total	32	1294.84					

Appendix IX

Comparison of the effects on skin spot development of disinfection with e.e.m.c., with Maneb (at 2 concentrations) and with boxing alone at different times of lifting.

Lifting Date	e.e.m.c.		Maneb (double normal concentration)		Maneb (normal concentration)		Boxing alone	
	SII	EII	SII	EII	SII	EII	SII	EII
6 Sept.	0.4	4.4	1.4	18.2	1.2	18.5	1.7	22.8
20 Sept.	0.2	1.7	2.1	24.3	2.2	21.8	2.3	28.3
4 Oct.	0.6	12.2	3.5	36.1	2.6	28.6	2.6	24.8
17 Oct.	1.4	17.4	2.6	27.9	4.2	36.8	6.0	38.5

Appendix X

Skin spot surface infection index in relation to different box storage treatments following clamp storage from lifting in early October, 1964-66.

Time from lifting (weeks)	1964			1965			1966	
	Boxed and disin-fected	Boxed and washed	Boxed only	Boxed and disin-fected	Boxed and washed	Boxed only	Boxed and disin-fected	Boxed only
- 4	-	-	-	0.6	2.1	2.1	-	-
- 3	-	-	-	-	-	-	-	-
- 2	-	-	-	0.8	4.0	3.0	0.2	2.1
- 1	1.0	4.7	5.7	-	-	-	-	-
0	2.2	6.0	6.8	0.7	3.9	3.8	0.6	2.6
+ 1	2.8	4.8	6.8	-	-	-	-	-
+ 2	-	-	-	1.3	1.9	4.3	-	-
+ 3	6.4	5.8	7.0	-	-	-	1.1	4.8
+ 4	-	-	-	2.0	6.0	4.1	-	-
+ 5	-	-	-	-	-	-	-	-
+ 6	5.3	6.8	7.7	2.1	7.6	8.8	1.3	6.9
+ 7	-	-	-	-	-	-	-	-
+ 8	-	-	-	-	-	-	-	-
+ 9	4.6	9.2	11.3	-	-	-	3.3	8.7
+10	-	-	-	4.3	8.5	6.7	-	-
+11	-	-	-	-	-	-	-	-
+12	5.3	9.3	13.2	-	-	-	4.4	9.6
+13	-	-	-	3.6	6.2	5.4	-	-

Appendix XI

Skin spot eye infection index in relation to different box storage treatments following clamp storage from lifting in early October 1964-66.

Time from lifting (weeks)	1964			1965			1966	
	Boxed and disin-fected	Boxed and washed	Boxed only	Boxed and disin-fected	Boxed and washed	Boxed only	Boxed and disin-fected	Boxed only
- 4	-	-	-	7.9	27.5	31.3	-	-
- 3	-	-	-	-	-	-	-	-
- 2	-	-	-	18.8	37.8	35.7	6.3	24.5
- 1	7.2	33.0	44.5	-	-	-	-	-
0	20.6	42.7	56.5	13.6	40.0	40.5	12.2	24.8
+ 1	20.6	30.4	41.3	-	-	-	-	-
+ 2	-	-	-	21.1	33.5	44.4	-	-
+ 3	39.8	40.8	44.9	-	-	-	14.2	32.7
+ 4	-	-	-	23.3	41.9	45.1	-	-
+ 5	-	-	-	-	-	-	-	-
+ 6	36.9	47.2	49.8	24.1	44.5	48.1	12.0	42.5
+ 7	-	-	-	-	-	-	-	-
+ 8	-	-	-	-	-	-	-	-
+ 9	26.7	46.4	57.1	-	-	-	22.1	41.4
+10	-	-	-	45.0	49.0	37.3	-	-
+11	-	-	-	-	-	-	-	-
+12	40.1	55.2	59.9	-	-	-	27.6	44.0
+13	-	-	-	38.3	44.0	43.8	-	-

Appendix XII

The effects of boxing treatments at varying periods after harvest and clamp storage on skin spot surface infection - 1964-1966.

Note: Arcsin transformation made on surface infection index data.

a) Analysis of variance - 1964

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	2	2.10					
Boxing treatment (T)	2	364.08	182.04	46.10***	3.23	5.18	8.25
Period in clamp storage treatment (L)	6	322.85	53.81	13.92***	2.34	3.29	4.73
(L x T) interaction	12	105.37	8.78	2.27*	2.00	2.99	3.64
Error	40	154.75	3.87				
Total	62	949.15					

b) Analysis of variance - 1965

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	1	0.47					
Boxing treatment (T)	2	261.94	130.97	82.89***	3.40	5.61	9.34
Period in clamp storage treatment (L)	7	306.64	43.81	27.73***	2.51	3.67	5.55
(L x T) interaction	14	60.73	4.34	2.74*	2.18	3.03	4.39
Error	24	37.94	1.58				
Total	48	667.72					

Appendix XII (contd.)

c) Analysis of variance - 1966

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	2	0.90					
Boxing treatment (T)	1	361.06	361.06	196.2***	4.28	7.88	14.19
Period in clamp storage treatment (L)	5	435.81	87.16	47.4***	2.64	3.94	5.98
(L x T) interaction	5	13.90	2.78	1.51	2.64	3.94	5.98
Error	23	42.42	1.84				
Total	36	855.09					

Appendix XIII

The effects of boxing treatments at varying periods after harvest and clamp storage on skin spot eye infection - 1964-1966.

a) Analysis of Variance - 1964

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	2	44.49					
Boxing treatment (T)	2	2296.83	1148.41	82.8***	3.23	5.18	8.25
Period in clamp storage treatment (L)	6	1471.81	245.31	17.72***	2.34	3.29	4.73
(L x T) interaction	12	680.02	56.67	4.10***	2.00	2.99	3.64
Error	40	552.40	13.81				
Total	62	5045.55					

b) Analysis of Variance - 1965

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	1	24.89					
Boxing treatment (T)	2	2760.02	1380.01	63.2***	3.40	5.61	9.34
Period in clamp storage treatment (L)	7	2076.94	296.70	13.62***	2.51	3.67	5.55
(L x T) interaction	14	1095.17	78.23	3.58**	2.18	3.03	4.39
Error	24	523.53	21.81				
Total	47	6480.56					

Appendix XIII (contd.)

c) Analysis of Variance - 1966

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	2	0.59					
Boxing treatment (T)	1	3336.98	3336.98	94.7***	4.28	7.88	14.19
Period in clamp storage treatment (L)	5	1816.64	363.33	10.20***	2.64	3.94	5.98
(L x T) interaction	5	268.55	53.71	1.51	2.64	3.94	5.98
Error	23	818.75	35.63				
Total	35	6241.53					

Appendix XIV

Maximum and minimum temperatures (°C) in clamp and insulated shed storage - 1964-67.

Month scale	Period from harvest (weeks)	1964-65		1965-66				1966-67			
		Store		Store		Clamp		Store		Clamp	
		Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.
Sept.	- 2							14	12		
	- 1			17	10			17	14		
	0	6	4	14	11			13	9		
	1	10	3	15	10			11	11		
Oct.	2	12	5	11	8	12	6	11	11	10	3
	3	10	4	11	7			11	9	12	8
	4	7	5	13	8	13	3	9	6		
	5	9	-1	10	6			7	5		
Nov.	6	9	3	9	4	7	1	7	6	8	1
	7	11	7	7	2			7	4		
	8	4	0	3	1	2	0	7	4		
	9	7	2	1	0			6	-1	6	-1
Dec.	10	4	2	4	2	6	1	5	3		
	11	5	2	7	3			6	3		
	12	4	1	4	2			6	3	11	-2
	13	8	4	4	2	9	1	4	2		
Jan.	14	6	3	5	3			4	2		
	15	4	2	6	2			4	2		
	16	3	2	2	1	7	0	5	4	11	-1
	17	6	2	4	2			6	3		
	18	6	3	6	4	8	1	8	6		
Feb.	19	6	3	4	1			9.	8		
	20	4	1	2	1	-	-1	5	3	8	0
	21	6	2	6	2			5	3		
	22	7	6	7	6			6	3		
Mar.	23	6	3	8	6			7	5		
	24			9	6			7	4		

Appendix XV

The average depths of the different levels of sporulation (mm.) and the total average depth of penetration of *Oospora pustulans* (mm.) into the tissue of tubers after various periods of clamp storage from lifting - 4 October 1966.

Time from lifting (weeks)	Dense	Sparse	Infrequent	Total
6	0.00	0.18	0.13	0.31
9	0.29	0.21	0.04	0.54
12	0.08	0.41	0.0	0.44
17	0.24	0.37	0.0	0.61
20	0.42	0.39	0.18	0.99

Appendix XVI

Skin spot development in tubers subjected to different box storage treatments after various periods of clamp storage from lifting - 4 October 1966.

Time from lifting (weeks)	Boxed and disinfected		Boxed only	
	SII	EII	SII	EII
6	1.3	12.0	4.8	42.5
9	3.3	22.1	6.9	41.4
12	4.4	27.6	8.7	44.0
-	-	-	Clamped	
20	-	-	5.9	33.0

Appendix XVII

Skin spot development related to different soil watering regimes - 1966.

Water (ins.) applied between 5 Sept. and 1 Nov.	Plots wet until 5 Sept. (8.17 in. water applied from 4 July)		Plots dry until 5 Sept. (1.66 in. water applied from 4 July)	
	SII	EII	SII	EII
0.0	4.6	26.2	-	-
0.5	4.0	21.2	2.7	13.9
1.0	5.0	31.0	2.8	15.2
1.5	3.2	14.9	3.0	16.8
2.0	2.3	16.0	3.7	23.5
2.5	2.4	17.1	3.6	23.5
3.0	2.3	13.0	3.9	27.1
3.5	2.0	11.3	4.5	20.3
4.0	-	-	4.5	28.2

Appendix XVIII

Effect of different watering regimes over the lifting period on skin spot development.

Comparison of treatment means using Students 't' test

Treatment before 5 Sept.	Water applied between 5 Sept. and 1 Nov. (in.)	Surface or eye infection	Number in sample	Mean	Arcsin transformation mean	Difference of means	S.E. of difference	t	Probability at t significant
Wet	1.0	Surface	4	5.0	51.82	4.45	1.18	3.77	0.01
	3.0		4	2.3	33.97				
Wet	1.0	Eye	4	31.0	-	19.7	6.95	2.83	0.05
	3.5		4	11.3					
Dry	0.5	Surface	4	2.7	8.84	3.02	2.21	1.36	-
	4.0		4	4.5	11.86				
Dry	0.5	Eye	4	13.9	-	14.3	9.14	1.57	-
	4.0		4	28.2					

Appendix XIX

The effects of different temperature and storage conditions on skin spot development.

Analysis of variance of surface infection (arcsin transformation)

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Treatments	3	3265.13	1088.3	77.34***	2.76	4.13	6.17
Error	60	844.47	14.1				
Total	63	4109.60					

Analysis of variance of eye infection

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Treatments	3	46301	15433	148.39***	2.76	4.13	6.17
Error	60	6280	104				
Total	63	52581					

Appendix XX

Skin spot development in tubers subjected to changing temperature conditions in later stages of storage.

a) Analysis of variance of surface infection (arcsin transformation)

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Treatments	3	218.45	72.81	4.45*	2.95	4.57	7.19
Error	28	458.35	16.36				
Total	31	676.81					

b) Analysis of variance of eye infection

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Treatments	3	1976.19	658.73	4.37*	2.95	4.57	7.19
Error	28	4213.19	150.47				
Total	31	6189.38					

Appendix XXI

Microscopic eye tests, made at 4 week intervals during storage, of tubers, naturally infected and artificially inoculated and subjected to different humidity and temperature regimes.

a) Storage temperature 5°C

Humidity level	Time from lifting (weeks)	Artificial Inoculation				Natural Infection			
		Eyes alive (per cent)		Eyes dead (per cent)		Eyes alive (per cent)		Eyes dead (per cent)	
		Fungus present	Fungus absent	Fungus present	Fungus absent	Fungus present	Fungus absent	Fungus present	Fungus absent
Damp	0	17	62	15	6	17	62	15	6
	4	37	43	18	2	21	49	25	5
	8	32	35	32	1	31	40	28	1
	12	32	32	35	1	17	44	37	2
	16	17	20	60	3	9	32	56	3
	20	19	14	66	1	11	24	62	3
Dry	0	17	62	15	6	17	62	15	6
	4	33	45	22	0	16	53	27	4
	8	27	45	25	3	18	52	23	7
	12	20	49	26	5	24	45	25	6
	16	16	17	62	5	15	22	59	4
	20	13	33	49	4	16	41	41	2

Appendix XXI (contd.)

b) Storage temperature 10°C

Humidity level	Time from lifting (weeks)	Artificial Inoculation				Natural Infection			
		Eyes alive (per cent)		Eyes dead (per cent)		Eyes alive (per cent)		Eyes dead (per cent)	
		Fungus present	Fungus absent	Fungus present	Fungus absent	Fungus present	Fungus absent	Fungus present	Fungus absent
Damp	0	17	62	15	6	17	62	15	6
	4	31	57	7	5	27	58	11	4
	8	45	37	16	2	25	53	22	0
	12	37	49	9	5	26	56	12	6
	16	38	44	16	7	37	41	17	5
	20	35	50	12	3	38	45	14	3
Dry	0	17	62	15	6	17	62	15	6
	4	29	55	14	2	24	67	8	1
	8	19	62	18	1	30	62	4	4
	12	22	69	8	1	18	61	13	8
	16	23	59	17	1	25	57	14	4
	20	37	52	8	3	29	61	9	1

Appendix XXII

The average depths of the different levels of sporulation (mm.) and the total average depth of penetration of Oospora pustulans (mm.) into the tissue of tubers, naturally and artificially inoculated and subjected to different humidity and temperature regimes.

a) Storage temperature 5°C

Humidity level	Time from lifting (weeks)	Artificial inoculation				Natural infection			
		Dense	Sparse	Infrequent	Total	Dense	Sparse	Infrequent	Total
Damp	0	0.00	0.09	0.08	0.17	0.00	0.09	0.08	0.17
	4	0.56	0.33	0.00	0.89	0.27	0.24	0.00	0.51
	8	0.34	0.25	0.07	0.66	0.11	0.31	0.03	0.45
	12	0.34	0.21	0.11	0.66	0.20	0.38	0.16	0.74
	16	0.25	0.45	0.00	0.71	0.26	0.50	0.00	0.76
	20	0.31	0.27	0.00	0.58	0.35	0.38	0.00	0.72
Dry	0	0.00	0.09	0.08	0.17	0.00	0.09	0.08	0.17
	4	0.36	0.32	0.02	0.70	0.39	0.17	0.10	0.66
	8	0.12	0.45	0.00	0.60	0.18	0.29	0.08	0.55
	12	0.42	0.24	0.03	0.70	0.36	0.18	0.06	0.60
	16	0.39	0.21	0.01	0.62	0.59	0.15	0.04	0.78
	20	0.40	0.30	0.00	0.69	0.48	0.32	0.00	0.80

Appendix XXII (contd.)

b) Storage temperature 10°C

Humidity level	Time from lifting (weeks)	Artificial inoculation				Natural infection			
		Dense	Sparse	Infrequent	Total	Dense	Sparse	Infrequent	Total
Damp	0	0.00	0.09	0.08	0.17	0.00	0.09	0.08	0.17
	4	0.00	0.54	0.00	0.54	0.07	0.54	0.00	0.01
	8	0.00	0.47	0.20	0.67	0.00	0.42	0.04	0.56
	12	0.12	0.46	0.09	0.67	0.00	0.09	0.27	0.36
	16	0.30	0.27	0.12	0.64	0.11	0.51	0.00	0.62
	20	0.04	0.43	0.00	0.47	0.00	0.38	0.09	0.47
Dry	0	0.00	0.09	0.08	0.17	0.00	0.09	0.08	0.17
	4	0.04	0.30	0.20	0.55	0.04	0.49	0.06	0.60
	8	0.00	0.39	0.11	0.50	0.05	0.41	0.09	0.56
	12	0.16	0.30	0.06	0.52	0.21	0.23	0.12	0.55
	16	0.31	0.31	0.12	0.74	0.11	0.37	0.00	0.48
	20	0.09	0.54	0.04	0.72	0.09	0.33	0.03	0.46

Appendix XXIII

Effects of different storage temperatures on the depth of penetration of *Oospora pustulans* into tuber tissue.

Comparison of means using Students 't' test

Temperature treatment	No. in sample	Mean depth of penetration (mm.)	Difference of means	S.E. of difference	t	Probability at t significant
5°C	20	0.668	0.102	0.103	1.00	-
10°C	20	0.566				

Appendix XXIV

Effects of delayed boxing and disinfection treatments on the rate of plant emergence and yield of the subsequent crop - 1965 and 1966.

a) Analysis of variance of rate of emergence 1965

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	40.75					
Treatments	3	88.76	29.6	3.01	3.86	6.99	13.90
Error	9	88.29	9.8				
Total	15	217.80					

b) Analysis of variance of rate of emergence 1966

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	49.18					
Treatments	3	56.59	18.86	2.89	3.86	6.99	13.90
Error	9	58.60	6.51				
Total	15	164.37					

Appendix XXIV (contd.)

c) Analysis of variance of yield 1965

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	77.42					
Treatment	3	7.30	2.43	0.3	3.86	6.99	13.90
Error	9	65.27	7.26				
Total	15	149.99					

d) Analysis of Variance of yield 1966

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	12.87					
Treatment	3	46.50	15.50	1.87	3.86	6.99	13.90
Error	9	74.38	8.26				
Total	15	133.75					

Appendix XXV

Effects of delayed boxing and disinfection treatments on the skin spot development of the resulting crop - 1965 and 1966.

a) Analysis of variance of surface infection 1965 (arcsin transformation)

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	102.24					
Treatment	3	28.56	9.52	0.75	4.35	9.55	18.77
Error	7	90.45	12.90				
Total	13	221.25					

b) Analysis of variance of surface infection 1966 (arcsin transformation)

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	33.25					
Treatment	3	105.30	35.10	6.88*	3.86	6.99	13.90
Error	9	45.94	5.10				
Total	15	184.49					

Appendix XXV (contd.)

c) Analysis of variance of eye infection 1965

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	202.59					
Treatment	3	853.95	275.0	15.6**	4.35	9.55	18.77
Error	7	123.11	17.58				
Total	13	1179.65					

d) Analysis of variance of eye infection 1966

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	469.50					
Treatment	3	1635.53	545.17	9.16**	3.86	6.99	13.90
Error	9	535.21	59.46				
Total	15	2640.24					

Appendix XXVI

Effects of disinfection treatments of seed tubers with different fungicides at lifting time on rate of plant emergence, yield and skin spot development in the resulting crop.

a) Analysis of variance of rate of emergence

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	47.55					
Treatment	2	32.38	16.19	2.82	5.14	10.92	27.00
Error	6	34.44	5.73				
Total	11	114.37					

b) Analysis of variance of yield

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	743.41					
Treatment	2	32.00	16.00	3.44	5.14	10.92	27.00
Error	6	27.84	4.65				
Total	11	803.25					

Appendix XXVI (contd.)

c) Analysis of variance of surface infection (arcsin transformation)

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	3.21					
Treatment	2	34.18	17.09	2.49	5.14	10.92	27.00
Error	6	41.08	6.86				
Total	11	78.47					

d) Analysis of variance of eye infection

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	91.42					
Treatment	2	391.26	195.63	4.93	5.14	10.92	27.00
Error	6	238.38	39.70				
Total	11	721.06					

Appendix XXVII

Effects of disinfection at planting of tubers with different levels of skin spot infection on plant emergence and skin spot development in the resulting crop.

a) Analysis of variance of rate of emergence

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	22.00					
Level of seed infection treatment (I)	1	17.85	17.85	6.23*	5.12	10.56	21.04
Disinfection treatment (D)	1	21.85	21.85	7.62*	5.12	10.56	21.04
(I x D) interaction	1	0.27	0.27	0.09	5.12	10.56	21.04
Error	9	47.90	2.87				
Total	15	87.60					

b) Analysis of variance of surface infection (arcsin transformation)

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	74.12	16.5				
Level of seed infection treatment (I)	1	16.56	16.56	3.80	5.12	10.56	21.04
Disinfection treatment (D)	1	62.17	62.17	** 14.2	5.12	10.56	21.04
(I x D) interaction	1	0.30	0.30	0.07	5.12	10.56	21.04
Error	9	39.37	4.37				
Total	15	192.53					

c) Analysis of variance of eye infection

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	1080.73					
Level of seed infection treatment (I)	1	1759.8	1759.8	** 12.75	5.12	10.56	21.04
Disinfection treatment (D)	1	2030.5	2030.5	** 14.7	5.12	10.56	21.04
(I x D) interaction	1	0.1	0.1	0.0	5.12	10.56	21.04
Error	9	2399.2	138.05				
Total	15	6189.49					

Appendix XXVIII

Effects of sprouting tubers at different temperatures on plant emergence, yield and skin spot development in the resulting crop.

a) Analysis of variance of rate of emergence

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Treatment	2	459.07	229.52	115.0***	4.26	8.02	16.39
Error	9	17.92	1.99				
Total	11	476.99					

b) Analysis of variance of yield

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	31.59					
Treatment	2	32.18	16.09	0.86	5.14	10.92	21.69
Error	6	112.66	18.8				
Total	11	176.43					

c) Analysis of variance of surface infection (arcsin transformation)

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	18.89					
Treatment	2	0.68	0.68	0.03	5.14	10.92	21.69
Error	6	113.58	18.9				
Total	11	133.06					

d) Analysis of variance of eye infection

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Replication	3	114.51					
Treatment	2	280.57	170.27	2.78	5.14	10.92	21.69
Error	6	411.25	68.5				
Total	11	806.33					

Appendix XXVIX

The effects on rate of plant emergence and skin spot development in the resulting crop of different levels of skin spot infection on seed tubers planted on two different dates - 1964.

Comparison number	Seed disease level	Planting date	Emergence rate or skin spot infection	No. in sample	Mean	Arcsin transformation mean	Difference of means	S.E. of difference	t	Probability at t significant
a	Free	16 April	Emergence rate	4	45.4	-	2.9	2.14	1.35	-
		30 April		4	48.3					
b	Slight, no eyes	30 April	Emergence rate	4	53.8	-	17.0	4.48	3.78	0.01
	Mod. or sev., all eyes			4	70.8					
c	Slight, some eyes	16 April	Surface infection	4	11.92	20.12	3.15	2.21	1.43	-
		30 April		4	15.85	23.27				
d	Free	-	Surface infection	8	7.25	15.55	6.14	1.36	4.52	0.001
	Slight, some eyes			8	13.88	21.64				
e	Free	30 April	Eye infection	4	45.8	-	26.0	5.35	4.86	0.01
	Slight, no eyes	16 April		4	71.8					

Comparison of treatment means using Students 't' test

Appendix XXX

The effects on rate of plant emergence and skin spot development in the resulting crop of different levels of skin spot infection on seed tubers of 2 different varieties planted on 2 soil types - 1965.

Comparison of treatment means using Students 't' test.

Comparison number	Seed disease level	Variety	Soil type	Emergence rate or skin spot infection	No. in sample	Mean	Arcsin transformation mean	Difference in means	S.E. of difference	t	Probability at t significant
a	Slight, no eyes	King Edward	Sandy	Emergence rate	4	47.8	-	8.0	2.96	2.71	0.05
			Med-loam		4	55.8					
b	Slight, some eyes	King Edward	Med-loam	Emergence rate	4	60.3	-	5.7	2.14	2.66	0.05
		Kerrs Pink			4	54.1					
c	Slight, some eyes	King Edward	Sandy	Surface infection	4	4.5	11.34	7.44	1.38	5.38	0.01
			Med-loam		4	10.5	18.78				
d	Free	King Edward	Sandy	Surface infection	3	3.0	9.55	4.74	2.88	1.65	-
			Med-loam		4	6.5	14.29				
e	Free	Kerrs Pink	Med-loam	Eye infection	4	35.7	-	10.2	5.65	1.81	-
	Slight, some eyes				4	45.9					

Appendix XXXI

The effects on plant emergence and skin spot development in the resulting crop of different levels of skin spot infection on seed tubers of 4 varieties on 2 soil types - 1966.

a) Analysis of variance of emergence rate

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Variety (V)	3	527.78	175.92	15.36***	2.84	4.31	6.60
Soil type (S)	1	4804.22	4804.22	459.73***	4.08	7.31	12.61
Seed infection level (D)	1	1316.78	1316.78	126.00***	4.08	7.31	12.61
(V x S) interaction	3	27.60	9.20	0.90	2.84	4.31	6.60
(V x D) interaction	3	310.39	103.46	9.03***	2.84	4.31	6.60
(D x S) interaction	1	29.30	29.30	2.80	4.08	7.31	12.61
(V x D x S) interaction	3	11.88	533.13	10.45			
Error	48	521.25					
Total	63	7548.20					

Appendix XXXI (contd.)

b) Analysis of variance of surface infection (arcsin transformation)

Source	df	SS	Variance	F	Table reading of F		
					P = 0.05	P = 0.01	P = 0.001
Variety (V)	3	7.29	2.43	0.41	2.84	4.31	6.60
Soil type (S)	1	948.02	948.02	163.0***	4.08	7.31	12.61
Seed infection level (D)	1	321.39	321.39	54.6***	4.08	7.31	12.61
(V x S) interaction	3	38.64	12.88	2.19	2.84	4.31	6.60
(V x D) interaction	3	75.56	25.20	4.28**	2.84	4.31	6.60
(D x S) interaction	1	18.98	18.98	3.23	4.08	7.31	12.61
(V x D x S) interaction	3	12.34	4.22	0.70	2.84	4.31	6.60
Error	48	292.68	6.08				
Total	63	1314.73					

Appendix XXXII

Mean value of the total sugar content of tubers in 3 temperature regimes of each of 5 varieties lifted mature and immature, measured at monthly intervals during storage (mgs./100 g. fresh wt.).

Variety	Tuber maturity at lifting	Total sugar content at monthly intervals					
		Sept.	Oct.	Nov.	Dec.	Jan.	Feb.
Kerrs Pink	Immature	248	977	640	800	985	-
	Mature	-	530	865	907	1142	612
King Edward	Immature	235	700	846	1208	1519	-
	Mature	-	285	522	827	849	1023
Arran Banner	Immature	436	1128	1000	1507	1314	-
	Mature	-	660	1265	1317	1300	1392
Arran Consul	Immature	409	786	1183	2049	1413	-
	Mature	-	681	1960	1626	1980	1206
Golden Wonder	Immature	400	984	1028	-	809	-
	Mature	-	675	1281	1025	777	1315

Appendix XXXIII

Mean value of the total sugar content of tubers in 5 varieties for each of 3 temperature regimes, after lifting mature and immature, measured at monthly intervals during storage (mgs./100 g. fresh wt.).

Temp. regime	Tuber maturity at lifting	Total sugar content at monthly intervals					
		Sept.	Oct.	Nov.	Dec.	Jan.	Feb.
10°C	Immature	345	476	305	313	182	-
	Mature	-	566	495	393	307	447
5°C	Immature	345	1826	1881	2382	2192	-
	Mature	-	566	1309	1756	1556	1786
Fluctuating (0-18°C)	Immature	345	697	632	1477	1250	-
	Mature	-	566	1732	1272	1766	1096