

**THE EFFECTS OF TWO FUNGICIDES ON STIGMA AND POLLEN
VIABILITY OF THREE STRAWBERRY CULTIVARS (*F x ananassa*)
AND THE IMPACT OF THESE ON FRUIT QUALITY**

**A thesis
presented to
the Faculty of the Graduate School
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Suggested Short Titles

EFFECTS OF FUNGICIDES ON STRAWBERRY
FRUIT QUALITY

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مجله مطلب

اثر دفع قیاح کش بر روی مادی و پرچم سه واریته توت زنی
(*E x ananassa*) و اهمیت آن بر روی وزن میوه و زیاده
میوه سی نارغوب

بیماری پوسیدگی خاکی توت زنی (*B. cinerea*) در حال حاضر توسط
استعمال قیاح کش که کنترل میشود. عمل واقعی این گونه مواد شیمیایی در دفع قیاح
نمودن اسپر قیاح از حیوان زن میباشد. در سالهای قبل آفات تنهات نشان داده
است که قیاح کش که نه تنها از پدیدار شدن بیماری حبه گری میماند بلکه قادر هستند که از
رشد دانه کرده درختان میوه حبه گری نموده که این عمل باعث غنیمت ماندن تخمدان گله و تنه کش
غیش تدبیری میوه میشود.

شرح ذیل تحقیقاتی است که در طول مدت دوسال از اول ژانویه ۱۹۸۱
تا ژانویه ۱۹۸۳ در دانشکده کشاورزی ملک فوالد و البته به دانشگاه ملک گیل انجام گرفته است.
هدف از این تحقیقات بررسی و مطالعه اثر معکوس قیاح کش بر روی محصولات کشاورزی و
پی آید آنها بر روی وزن و شکل میوه بود. چهار آزمایش مختلف به قرار زیر انجام شده
۱- در تابستان سال ۱۹۸۱ سه نوع واریته توت زنی آفتاب و دفع قیاح در دوطرف مختلف کمار بردند.
۲- در پاییز سال ۱۹۸۱ آزمایش قبی در داخل گلخانه مکرر گردید و قبل از استعمال قیاح کش پرچم طبعی گله مبر گردید.
۳- در زمستان سال ۸۲-۱۹۸۱، غنطت دفع قیاح کش قبی بر روی رشد دانه کرده توسط سیدو سیدو مطالعه شد.
۴- در تابستان سال ۱۹۸۳، آزمایش سال ۱۹۸۱ تدرار و مطالعه ختیری بر روی خصلت گره افشان صورت گرفت.
نتایج این آزمایشات نشان داده که در گلخانه و آفتابخانه استعمال قیاح کش میتواند از رشد
دانه کرده و توسعه لوله گره حمایت نماید. قابل توجه است که شصت فوق مختلف قیاح گرفته شده
در زمزمه بوده زیرا که استعمال قیاح کش در گلخانه اثری بر روی وزن و شکل میوه ندارد. تفاوت
فوق میتواند علت حضور خصلت گره افشان در زمزمه باشد که باعث انتقال دانه کرده از گیاهی
به گیاه دیگر میشوند. در پایان چنین نتیجه گیری میشود که در صورت نبودن خصلت گره افشان
و شش طبعی استعمال قیاح کش باعث کم شدن وزن میوه و زیاده میوه سی
نارغوب میشود.

ABSTRACT

M.Sc.

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Plant Science

THE EFFECTS OF TWO FUNGICIDES ON STIGMA AND POLLEN VIABILITY OF THREE STRAWBERRY CULTIVARS (*F x ananassa*) AND THE IMPACT OF THESE ON FRUIT QUALITY

The control of *Botrytis cinerea* on strawberries depends upon the application of protective fungicides. Their mode of action is to prevent the fungal spore from germinating, to prevent penetration by the fungal germ tube, or to prevent mycelial growth.

Fungicides can reduce pollen germination and the length of the pollen tube.

The investigation was conducted to study the adverse effects of fungicides on strawberry pollen and stigma viability and the impact of these on fruit quality and yield.

Four experiments were undertaken:

- I- A field experiment using three cultivars (Redcoat, Sparkle and Bounty) and two fungicides (Captan and Easout) applied at early or late bloom and two different levels of pollination (caged and uncaged).
- II- A greenhouse experiment using the same materials, in which all flowers were emasculated to control pollination.
- III-A laboratory experiment to evaluate the critical concentration of two fungicides (Captan and Easout) with respect to percent pollen germination and length of pollen tube using 0, 500, 1000 and 2000ppm.
- IV- A field trial to confirm the results obtained in I.

Application of either fungicide reduced pollen germination and tube length, however this effect was not important in the field because of the transfer of viable pollen to treated flowers by pollinating insects.

In some situations, where plants were isolated and no insects were present, the application of fungicides during the flowering period reduced yield and berry quality.

RESUMÉ

M.Sc.

Shahrokh Khanizadeh

Phytotechnie

L'effet de l'application de deux Fongicides sur les Stigmas et la Viabilité du Pollen et sur la Qualité des Fruits Produits de Trois Cultivars de Fraise (F x ananassa)

L'application de fongicide peut contrôler Botrytis cinerea dans les fraisiers. Les différents modes d'action de ces fongicides sont soit l'inhibition de la germination des spores, soit l'inhibition de la pénétration du tube germinatif, soit l'inhibition de la croissance de mycelium.

Les fongicides peuvent aussi réduire la germination du pollen ou la longueur du tube pollinique.

Quatre expériences ont été conduites afin d'étudier les effets des fongicides sur la viabilité du pollen et des stigma des fraisiers et leurs influences sur le rendement qualitatif et quantitatif des fraisiers.

Les quatre expériences sont:

- I -Un essai de parcelles dont les facteurs étudiés étaient trois cultivars (Redcoat, Sparkle et Bounty), deux fongicides (Captan et Easout), deux temps d'application (au début et à la fin de la floraison) et deux modes de pollinisation.
- II -Une expérience en serre où les mêmes facteurs furent étudiés sauf que dans cette expérience les fleurs furent emasculées afin de contrôler la pollinisation.
- III -Une expérience en laboratoire où la germination du pollen et la longueur du tube pollinique furent mesurées à quatre niveaux de concentration, 0, 500, 1000 et 2000 ppm, afin d'évaluer la concentration critiques des deux mêmes fongicides.
- IV -Un essai en plein champ pour vérifier les résultats obtenus en I.

L'application des deux fongicides réduit la germination du pollen et la longueur du tube pollinique. Néanmoins ces effets ne sont pas importants en plein champs à cause de l'apport de pollen viable sur les fleurs traitées par les insectes pollinisateurs.

Cependant dans certains cas où les fraisiers sont isolés et où la présence d'insectes pollinisateurs est négligeable, l'application de fongicide à la floraison peut réduire le rendement et la qualité des fruits.

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AUTO-BIOGRAPHICAL SKETCH

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In 1981, he began his graduate work in the Department of Plant Science. At that time, he received a teaching assistantship in the Department of Plant Science, and later one from the Department of Physics & Chemistry. He continued with this work until the end of his studies.

In December 1983, after two years, the author completed his Masters studies.

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LIST OF STANDARD ABBREVIATIONS

Abbreviations	Definitions
ml	millilitre
l	litre
g	gram
mg	milligram
mm	millimetre
kg	kilogram
LD ₅₀	median lethal dose
cv	cultivar
°C	degrees centigrade
T	metric tonne
DF	degrees of freedom
C.V.	Coefficient of Variability
LSD	Least Significant Difference
MS	mean square
WP	wettable powder

1. INTRODUCTION

Fruit rots are probably among the most important diseases of strawberries (Plakidas,1965; Gourley,1968; Eaton and Chen,1969a; Mass,1978a), grey mould (Botrytis cinerea Pers.) is one of these. Losses of strawberries due to fruit rots vary from year to year and from locality to locality, depending on weather conditions. Infections may be severe during wet weather when 30% to 50% of a strawberry crop may be lost (Mass,1978a, Mass,1978b). Relatively speaking, B. cinerea Pers. is a cool weather disease and is common in northern temperate regions (Plakidas,1965).

B. cinerea Pers. may attack various parts of the strawberry flower soon after the buds open (Jarvis and Borecka, 1968) and conidia can germinate between 0°C and 26°C (Schneider and Orelli,1912; Brooks and Cooley,1917; Doran,1922). However,the growth rate is optimal at about 20-22°C and decreases markedly above 25°C (Jarvis,1977).

The pathogen usually lives for a time as a saprophyte on dead or dying petals,sepals and stamens before invading the base of the swollen receptacle (Powelson,1960; Bhatt and Vaughan, 1962). The presence of pollen grains affects spore germination (Brown,1922; Chou and Preece,1968) and is usually associated with increased infection (Ogawa and English,1960). Such an increase has been noted on strawberry flowers and fruit (Jarvis and Borecka,1968; Chou and Preece,1968; Borecka and Rudnicki,1969).

The control of B. cinerea by a fungicide depends on delivering the chemical to the site of infection at the proper time (Brandes, 1971). Therefore, fungicidal sprays are applied during the blossoming period at intervals of 8-10 days and a final application is made approximately 2 weeks before harvest (Bhatt and Vaughan, 1962; Freeman, 1964; Eaton and Chen, 1969a; Macswan, 1982). This prevents B. cinerea from becoming established as a saprophyte on the senile or dead floral organs, from where it could invade the fruit (Bhatt and Vaughan, 1962).

For many years, there has been speculation as to the possible adverse effects of fungicidal sprays applied at blossom time on pollen germination and fruit set in many crops. These crops include apples (Rich, 1957; Eaton, 1963; Church and Williams, 1977; Church and Williams, 1983; Church and Morgan *et al.*, 1983; Church and Cooke *et al.*, 1983), cherries (Eaton, 1961) and cranberries (Shawa, Doughty and Johnson, 1966) etc., as well as strawberries (Eaton and Chen, 1969a). In all cases most fungicides inhibit pollen germination *in vitro* when added to the medium (Rich, 1957; Eaton, 1961; Gartal, 1961; Kwack and Macdonald, 1966; Cristoferi *et al.*, 1966; Shawa *et al.*, 1966; Eaton and Chen, 1969b; Dancs and Kiss, 1970; Burth and Ramson, 1974).

This study was undertaken to elucidate the effects of fungicide and time of application on stigma receptivity, pollen viability, fruit yield and quality. The role of pollinating insects in fruit development in the field was also investigated.

2. REVIEW OF LITERATURE

2.1. The Strawberry

The English word "strawberry" is thought to be derived from the Anglo Saxon word "streowberie" which relates to the spreading nature of the runners of the plant which are "strewed" (Fraser, 1926). The word was read as "strawed-berry". In the fifteenth century it became known as "strawberry" (Lidgate 15th century). The name, therefore, has no connection with straw (which is sometimes used for mulching the plant) but relates to the behaviour of the plant itself (Fraser, 1926).

Another conjecture is that the Anglo-Saxon word "streo" meant hay; according to one theory, the Anglo-Saxons in 900 A.D. called the strawberry "hayberry" because it ripened at the time the hay was mown (Darrow, 1966).

2.1.1. Early History of the Strawberry

Strawberries were known as wild plants at the time of the Greek and Roman empires(Fraser,1926). Pliny the Elder (23-79 A.D.) mentioned the strawberry in his treatise (Darrow,1966). By 1300 A.D., the strawberry was cultivated in Europe. At that time, Fragaria vesca was transplanted from the wild into gardens. The plant was esteemed for its flowers rather than for its fruit (Darrow,1966). At the close of the sixteenth century, the "wood strawberry" F. vesca was common in English gardens (Fletcher,1917). Until the end of seventeenth century, the wood strawberry was preferred by strawberry growers (Darrow,1966).

Gardeners were satisfied to grow the wild species and there had been no improvement in the European species. In fact, there was little progress until the introduction of two American species, Fragaria virginiana and Fragaria chiloensis when the first step in the domestication of the wild strawberry began (Fletcher,1917). In 1624, the Virginia strawberry (F. virginiana) was introduced to France from an American garden (Fraser,1926). In 1712, the journey of a French spy to Chile resulted in the introduction of F. chiloensis, the other American parent of Fragaria grandiflora (now known as

F x ananassa) our present day large-fruited strawberry (Fletcher,1917).

In about the year 1800; the pine strawberry (one of the cultivars

resulting from hybridization of the Chilean and Virginian species) was introduced from Europe to America (Fraser, 1926). In America, the pine strawberry approached perfection in fruit but the plants were poor, whereas native American plants were hardy and vigorous but the fruits were small and insipid. In 1834, the cultivar Hovey (the result of a cross between these two species) was developed. By 1850, it had established the strawberry as one of the leading fruits in America (Fletcher, 1917). In the words of Fraser (1926): "Much has been done in the development of cultivars and more will be done, but the advent of Hovey centered so much attention on the strawberry that the American public have [sic] never since lost interest in this fruit" (Fraser, 1926).

2.1.2. Botany and Morphology of the Strawberry

2.1.2.1. Description of the Genus *Fragaria*

"X=7, acaulescent, more or less hairy, perennial herbs with basal leaves and long liliform runners from the axils, which root and form new plants. Petioles mostly long and channeled above, stipules adnate at the base of the petiole, large, mostly scarious and brown, persistent and covering the rootstock. Leaves three foliate, or sometimes unequally imparinnate, i.e., with a pair of much smaller lateral leaflets below the normal ones, the lateral ones oblique, the inner half usually smaller. Scape mostly about as long as the petioles, cymosely branched; pedicels slender, erect when in flower, curved when in fruit" (Hedrick, 1925); (Fig. 1).

The flower buds are borne at the axil of leaves on the crown, and the inflorescence is cymose. The early central flowers open first and are much larger than the others (Fig. 1). The flowers are polygamous^{us} dioecious or hermaphrodite (Fig. 2). The male flowers are larger and showier. All the flowers are five parted and are often six to eight parted (Westwood, 1978).

Calyx lobes form a flat hypanthium, augmented by many shorter and mostly narrower outer calyx-lobes or bractlets.

There can be twenty or fewer stamens and some can be abortive.

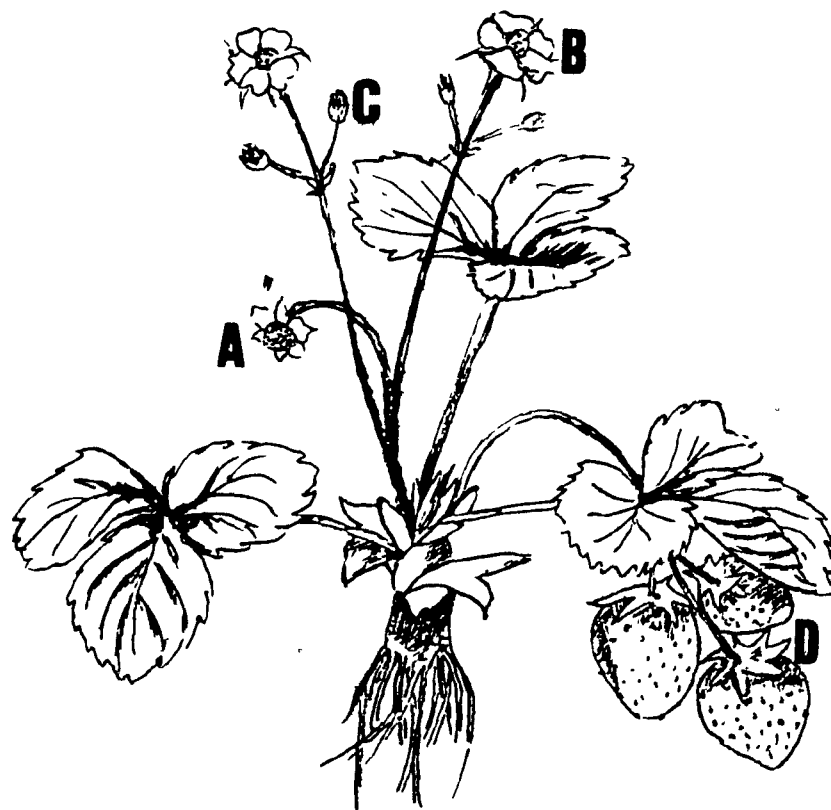


Figure 1. Strawberry plant.

- A) primary flower
- B) secondary flower
- C) tertiary flower
- D) primary and secondary fruits

Fig. 2. Strawberry flowers

- (A) Female flowers
- (B) Hermaphrodite



The filaments are usually shorter than the receptacle and the anthers are oblong (Fig. 3). The receptacle is either roundish or conical, bearing numerous pistils with lateral styles. At maturity, the receptacle becomes enlarged and juicy. This portion is the strawberry fruit (Hedrick, 1925; Westwood, 1978), (Fig. 4).

2.1.2.2. Distribution

Strawberries are found in almost every part of the world. they are cultivated in the temperate regions of Europe and Asia (Hedrick, 1925; Darrow, 1966). In the Americas, they occur from Alaska to Southern Chile on the Pacific Coast, for a thousand miles Northward up along the Atlantic Coast of South America and over much of North America, as well as on the mountain ranges of both countries (Hedrick, 1925; Fraser, 1926; Darrow, 1966). There is considerable variation among the types of strawberry plants owing to their remarkable ability to adapt to climate and soil (Hedrick, 1925). In cultivation, they are found in regions having a cool climate or grown during the cool months of the year (Fraser, 1926). Neither wild nor cultivated strawberries are esteemed in tropical countries. There, and in some sub-tropical countries, the plants do not bear abundantly, the fruits being deficient in size, colour, flavour and the delicate fragrance characteristic of certain cultivars when grown under favourable conditions (Fraser, 1926).

No plant better illustrates how a fruit can reach its highest

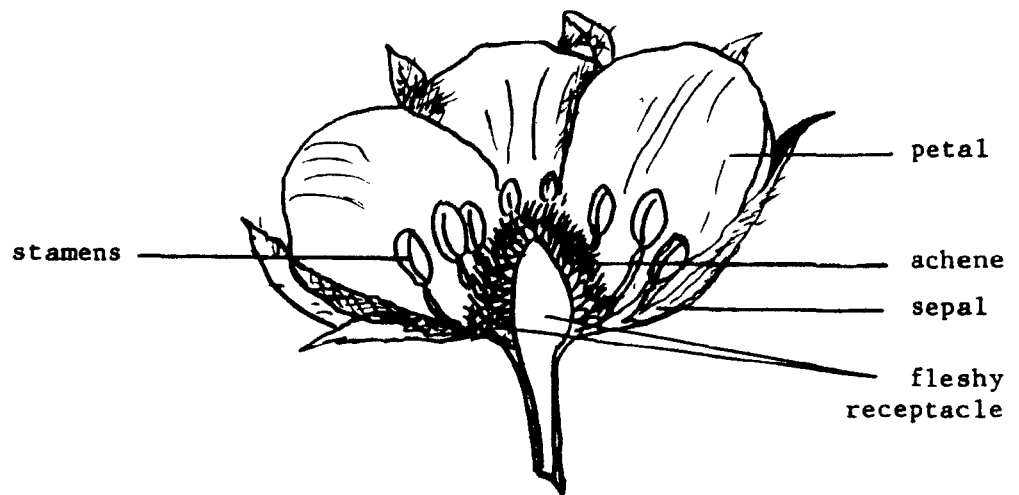


Figure 3. Median section through a flower of F x ananassa.

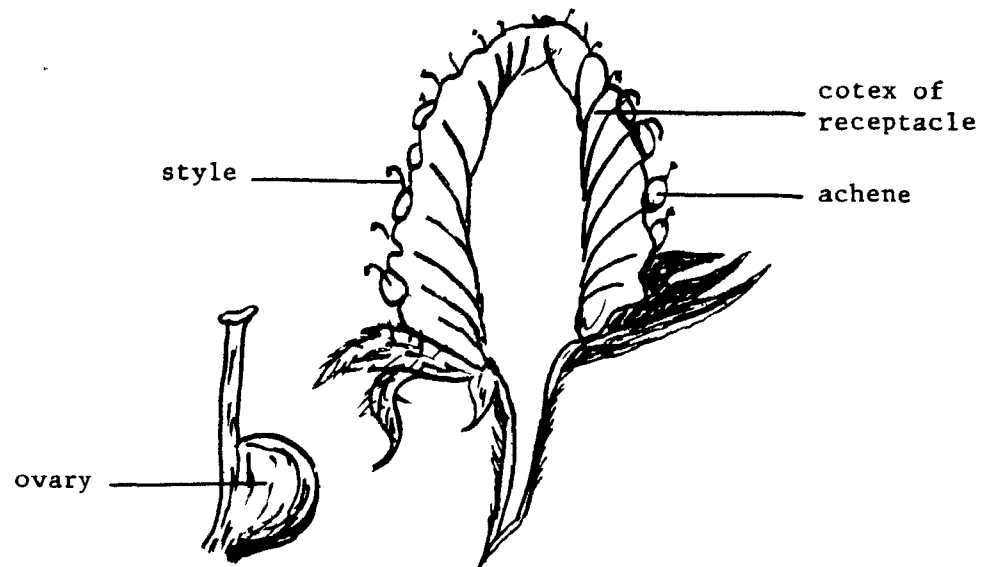


Figure 4. Median section through a fruit of F x ananassa

development in flavour and quality when allowed to grow until full maturity, at either the most northerly or most southerly points. Consequently, it is in Northern Europe, Northern America and at high elevations where these plants have been domesticated and they grow at their best. Incidentally, cultivation does not improve the flavour (Fraser,1926).

Currently, strawberry cultivars are divided into three groups according to their fruiting habit (Darrow,1966):

- 1) June bearing; most cultivars are of this type (short day plants),
- 2) Everbearing; the plants fruit two or more times per season (long day plants) and
- 3) Day neutral; in this group flowering is continuous from early spring to late in fall.

The most popular cultivars are the single crop or June bearing strawberries and the most important ones in the USA are: Northwest, Blakemore, Shasta, Headliner, Tennessee Beauty, Dixieland, Marshall, Sparkle, Robinson and Midway. Redcoat, Sparkle, Veestar, Vibrant, Grenadier and Bounty are grown extensively in Eastern Canada (Darrow,1966; Craig,1981; Craig,1982).

In 1977, the commercial acreage of strawberries in Canada and United State was about 10.129 ha (Shoemaker,1977). At the present time Canada and USA produce 360,187 T of strawberry (F.A.O., 1982).

2.1.3. Fruit Set and Development

In most angiosperms, the presence of fertilized ovules is essential to prevent abscission of the flower and to stimulate fruit development (Nitsch,1950; Nitsch,1955; Westwood,1978). Nitsch, in 1950 showed that the development of the strawberry receptacle was entirely dependent upon the presence of fertilized achenes (Fig. 5). This observation was confirmed by Tukey in 1952. Swelling of the strawberry receptacle is caused by growth substances which are active in the Avena test,a standard test for auxin (Nitsch,1950). This has led to the belief that the factor supplied by the seeds (achenes) is an auxin (Gustafon,1939; Nitch,1950) and the complete removal of fertilized achenes after pollination, or anything which causes abortion of achenes, will stop berry development. The result is reduced fruit weight and malformation of berries (Nitsch,1950).

In short, only fertilized achenes are active and the weight and shape of the fleshy part of a strawberry is a function of the number and distribution of developed achenes (Nitsch,1950).

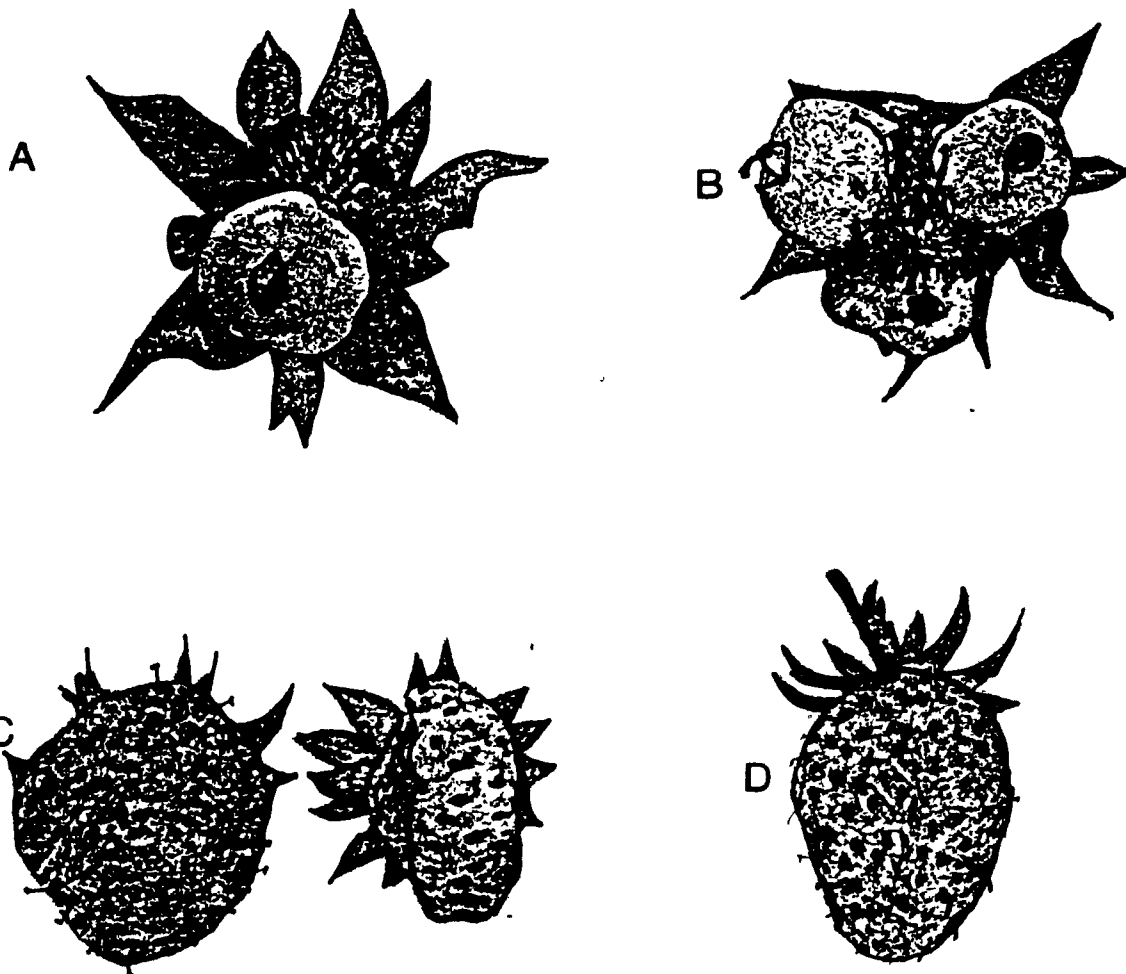


Figure 5. Illustration of the importance of the presence of fertilized achenes for the development of strawberry fruits.

- A) Growth of the strawberry receptacle induced by one single fertilized achene
- B) Growth induced by three fertilized achenes
- C) Growth induced by three rows of achenes
- D) Control

From: Nitch, 1950

mould disease". Fruit may become infected at any stage of development (Plakidas,1965; Laemmlen,1980).

Grey mould is both a saprophyte and a facultative parasite. It survives adverse climatic conditions (hot summer and cold winter) in the form of sclerotina and dormant mycelium in dead plant material. Under optimal conditions the fungus grows and sporulates on dead and moribund parts of strawberry plantings as a saprophyte. It is then spread by wind, rain and the fruit pickers. In moist conditions the conidia germinate in a few hours (Plakidas,1965). The pathogen usually lives for a time as a saprophyte on dead or dying petals and stamens, before invading the base of the swelling receptacle (Powelson, 1960).

Grey mould is a cool weather disease and is more common in cooler regions. Humidity, not temperature, is the most important of the environmental factors to influence the incidence and severity of the disease (Plakidas,1965). Plakidas (1965) showed that, in Louisiana, if a period of cloudy, rainy weather persists for about a week during harvesting, 70% to 75% of the fruit will rot in the field. If such a period occurs during the peak of harvesting, the growers there may lose a million dollars or more in a week. Powelson (1960) reported that B. cinerea caused over 90% of the fruit rot in the fields surveyed in British Columbia, Washington, Oregon and California in 1956, 1957, and 1958. In 1978b, Maas reported current statistics showing the value of losses of strawberry fruit, to pre- and post-harvest fruit rots,

conservatively estimated to be in the millions of dollars annually to growers, processors and retailers. It was also reported, that during wet weather, 30% to 50% or more of the strawberry crop may be lost to Botrytis rot even with liberal use of fungicides. In the last 20 years, much research effort has been directed toward controlling the disease, usually by repeated application of fungicides beginning at the blossom stage (Jarvis, 1962b; Jarvis and Borecka, 1968; Eaton and Chen, 1969a).

Studies of the biology of the pathogen and host have been made to enable fungicides to be used more effectively (Brown, 1922; Jarvis, 1961; Smith, 1900; Jarvis, 1962a & 1962b). It has also been reported that a number of fungi other than B. cinerea are associated with strawberries at bloom, green, and ripe fruit stages of development (Bhatt and Vaughan, 1963). Bhatt and Vaughan (1963) demonstrated that fungi isolated from strawberries showed various degrees of antagonism towards Botrytis spp, ranging from mutual inhibition to no effect on each other's growth. Dendrophoma obscurans and Pullularia pullulans, however, prevented growth of Botrytis cinerea on green and ripe strawberries (Bhatt, 1962; Bhatt and Vaughan, 1963). Furthermore, they suggested that in areas where D. obscurans occurs naturally on strawberry fruit, it undoubtedly prevents many infections by B. cinerea. It has been shown that spore suspensions of Penicillium, Pullularia, and Cladosporium herbarum either prevented or delayed infection by B.

cinerea, (Bhatt,1962). This suggested that fungicides not only prevent B. cinerea growth but may depress the growth of the other fungi which could naturally inhibit the growth of B. cinerea. It has been shown that spore suspensions of Penicillium, Pullularia, and Cladosporium herbarum either prevented or delayed infection by B. cinerea, (Bhatt,1962).

Many workers have suggested that if fungicidal sprays are applied during the blossoming period, at intervals of 8-10 days, and the last application is made approximately 2 weeks before harvest, this will completely prevent the incidence of B. cinerea (Powelson,1960; Bhatt and Vaughan,1963; Freeman,1964; Howard and Albregts,1982; MacSwan and Gashaira,1982; Moore and Bordelon,1982). The purpose of applying fungicides at intervals, is to prevent B. cinerea from becoming established as a saprophyte on the senile or dead floral organs. If it becomes established, B. cinerea can invade uninjured tissue (Powelson,1960; Bhatt and Vaughan,1963).

In 1960, Ogawa and English published a photo micrograph showing pollen grains apparently stimulating Botrytis spore germination and germ tube growth. They interpreted this as indicating that infection of petals was enhanced by the presence of pollen grains. Later it was reported that a heavy deposit of pollen grains might serve as a locus of infection of flower and other parts of American holly Ilex opaca Ait., by B. cinerea (Bachelder and Orton,1963).

These reports led Chou and Preece (1968) to examine B. cinerea conidia, pollen grains, petals and fruits of the F. ananassa cv. Bailey and Royal Sovereign. They reported an occasional apparent stimulation of fungal growth observed near pollen on leaves. Pollen could have effects on spore germination and development of B. cinerea infections on strawberry petals and fruits. It could also increase the speed and severity of B. cinerea attacks on strawberry fruit (Chou and Preece, 1968). This suggested that the presence of pollen had an effect on the early stage of infection by fungi.

In 1968, Jarvis and Borecka, found that among all six cultivars tested, susceptibility to infection was greatest at the open flower and white bud stage and least at the green bud stage. The calyx and corolla were more readily infected than the unripe receptacle. It was suggested that the calyx and corolla may later provide a base for infection of the healthy ripe receptacle, (Stevens,1922; Anderson,1946; Powell,1952; Stoddard and Miller,1956).

Powelson demonstrated, in 1960, that if the petals, stamens and calyxes were removed, under controlled conditions, severe disease developed less frequently. Field observation indicated that much of the fruit rot resulted from the spread of infection from sepals and petals to the fruit (Wilkinson,1954; Kirby,Moore and Wilson,1955; Stoddard and Miller,1956).

The above results are similar to the results of Powelson (1960),

Bhatt and Vaughan (1963) and Jarvis and Borecka (1968) who showed that the best time for application of fungicides to prevent B. cinerea is likely to be during blossoming. However, the primary effect of spraying at this time may be on the pollination process. This effect may be exerted on the dehiscence mechanism of the anthers. Affected anthers appear grey or orange rather than the usual yellow as has been shown on apples (Williams and Wilson, 1970). Williams and Wilson (1970) suggested that if fungicide must be applied during the flowering period of apple trees, it would be less damaging to do so just after full-bloom of the main crop cultivar rather than just before, as is often done. Several workers have reported adverse effects of fungicides on pollen and fruit quality of various species (Rich, 1957; Eaton, 1963; Shawa et al., 1966; Eaton and Chen, 1969a & 1969b; Gentile and Gallagher, 1972).

2.3. What a Fungicide is and How it Works

The fungi are a form of plant life containing no chlorophyll, therefore, they depend upon an organic source for nutrition and energy (Horsfall,1956). However, when the source of nutrition and energy is supplied by a commercial crop, fungus infection can become an economic problem and some form of fungus control should be employed.

Fungus can be killed by various means,such as ultraviolet light or by heat (Horsfall,1956). There are two basic principles of fungus control: protection and therapy. Protection refers to dealing with possible fungus attacks before they can enter the host. Therapy, on the other hand, refers to dealing with the fungus after it has entered the host (Horsfall,1956). In these experiments, protection was dealt with rather than therapy.

The word fungicide, meaning "to kill a fungus", is derived from the Latin words fungus and caedo.

Two different procedures are available to achieve protection with fungicides:

- a) The fungicide is used to seek out the fungus at rest (either before or after the host has come into contact with the fungus).
- b) The fungicide spray leaves a residue forming a layer on the surface of the host, awaiting the mobile fungus (Horsfall,1956).

2.3.1. Easout and Captan

2.3.1.1. Easout

Easout (Dimethyl 4,4'-O-phenylenebis 3-thioallophanate) is an organic compound used as a curative, preventative and systemic fungicide. The synthesis of Easout was reported in 1969 through the Nippon Soda Co., Ltd. of Japan. Later it was developed in the U.S. by Pennwalt Corporation, Mallinckrodt chemical works. Its mammalian toxicity (LD_{50}) is reported to be 7500 mg kg^{-1} . Easout is available as 50 or 70% WP. It acts as a curative or protectant against a wide range of fungal infections in agriculture. It has been reported that Easout is not phytotoxic when used as directed. It is worth mentioning that it is stable in sunlight, thus giving long residual control (Thomson, 1982 & 1983).

2.3.1.2. Captan

Captan (N- trichloromethylthio- 4 -cyclohexene-1, 2-dicarboximide) ranks second to the dithiocarbamates in importance as an organic fungicide in agriculture (Lawrence and Block, 1968). The synthesis of Captan was first reported in 1949 and it was registered in Canada in 1951 (Thomson, 1982 & 1983). It is also known under the names Merpan, Orthocide or Vondcaptan (Thomson, 1982 & 1983). Captan, 93-95% pure, is a yellow powder with a distinct odour. It is available in different formulations, mainly 3.5 7.5% dust or 50% and 80% WP (Thomson, 1982 & 1983). It is practically insoluble in organic solvents.

Its water-solubility is less than 0.5 ppm. It is fairly mobile in moist soils and aquatic systems (Ziraldó,1982). Its mammalian toxicity (LD_{50}) has been reported to be about 9000 mg kg^{-1} . It acts as a protectant against a wide range of fungal infections in agriculture. It is used as a foliar treatment and as a seed dressing (Thomson,1981 & 1982).

It is estimated that 13 million kilograms of Captan are used annually in the world. In Canada the use of Captan is around 300,000 kilograms annually. The range of uses and strengths employed are: 82% foliar and fruits, 10% seed treatment, 5% home and garden and 3% non-agricultural (Ziraldó,1982).

There have been reports about the use of Captan in agriculture; some have been controversial. So far, it indicates that Captan does not pose a mutagenic risk to humans, however, interest in the fungitoxicity of Captan has approached that of the dithiocarbamates and Oxine. Captan's toxicity has frequently been reviewed (Horsfall,1956; Rich,1960 & 1963; Lukens,1965; Eaton and Chen, 1969a & 1969b).

It has been reported that fungitoxic doses of Captan strongly inhibit respiration of fungi, presumably by inhibiting certain respiratory enzymes (Torgeson,1969). Captan may cause these inhibitions in fungal cells and these inhibitions may lead to cell death. So, it is not surprising that it might inhibit the respiration of pollen grains and consequently affects the fertilization process (which will be discussed in the next section).

2.4. The Adverse Effects of Fungicides on Strawberry Quality and Yield

Both organic and inorganic fungicides (some with the addition of nutrient elements such as calcium and boron) have been tested for control of grey mould of strawberries. The possible adverse effects of fungicide spray application at blossom-time, on pollen germination and fruit set, have been studied in many crops, including apple, cherry, cranberry, strawberry, etc. (Bhatt and Vaughan, 1962; Freeman, 1964; Eaton and Chen, 1969a; Tapio, 1972; Bristow, 1981; Shawa, 1981; Church and Williams, 1983; Church and cooke et al., 1983; Church and Morgan et al., 1983).

In most cases fungicides applied gave some control of grey mould (Freeman, 1964; Brandes, 1971; Tapio, 1972; MacSwan, 1980; Gourley and Delbridge, 1980; Abdel-Rahman, 1980; Howard and Albrechts, 1982; Moore and Bordelon, 1982; MacSwan, 1982). In 1971, Brandes found this to be true only if the fungicides were delivered to the site of infection at the proper time.

Rich (1957) reported that none of the fungicides he tested reduced pollen germination or fruit-set when applied to apple trees in bloom. He also found sufficient viable pollen following application of the fungicide Captan during the bloom stage. Further, he found that in vitro, apple pollen would not germinate in sucrose solutions containing

the fungicides. His work in vivo was based on a study of the application of apple pollen from unsprayed Cortland flowers to sprayed McIntosh flowers as well as pollen from sprayed flowers to unsprayed McIntosh flowers. In addition, his data showed that sulphur, Captan, Glyodin, Dichlone, and Ferbam, applied during the bloom stage reduced pollen germination of sprayed flowers, but he did not consider the reduction to be serious.

Church, Williams, Morgan, and Cooke (1978) reported laboratory experiments that showed fungicides when sprayed on open flowers may damage pollen. In order to relate those results to orchard practice apple trees, cv. Cox's Orange Pippin, were sprayed with fungicides during full bloom stage. Clusters which had been hand pollinated two hours before spraying were unaffected by the spray, but Captan and Dinocap reduced the fruit set of branches which had not been pollinated by hand.

In 1961, Eaton reported that among the fungicides he tested on sweet cherry Prunus avium L., sulphur did not reduce the germination of pollen whereas Dichlone and Ferbam significantly reduced germination. Captan at 2g l^{-1} almost entirely prevented pollen germination and arrested pollen tube elongation.

Powell (1954) and Freeman (1964) reported that strawberry plants had "benefitted nutritionally" from Captan and that fruit size was increased. Increase in fruit size might be due to the increased

respiration process reported after Captan application (Yor-ganchena, 1973). Eaton (1963) indicated that Captan sprays applied to anthers significantly reduced apple pollen germination, but not equally for all cultivars. His work confirmed the results of Schmidt (1956), Cristoferi et al., (1966) and Braun and Schonbeck (1965) who reported that cultivars or species may differ in their reactions to a fungicide. In addition, Eaton (1963) suggested that Captan inhibited the pollen tube elongation of some cultivars.

Evidence on the effect of fungicides on pollen in undehisced or recently dehisced anthers is conflicting (Dancs and Kiss, 1970; Rich, 1957; Shawa et al., 1966; Lockhart, 1967). However, it has been shown that some fungicides used in British orchards can reduce the germination of apple pollen on the stigma (Legge and Williams, 1975; Church and Williams, 1977).

In 1964 Freeman, reported that a single application of Captan gave a 37% increase in the yield of sound fruit and four similar sprays gave a 59% increase in sound fruit over the unsprayed plots. Further, he confirmed the desirability of a full spray schedule. He concluded that fruit size was affected by treatment and that plots treated with Captan produced larger fruit. Freeman's work (1964) confirmed the results of Powell (1954) who reported that strawberry plants benefitted nutritionally from Captan. He also reported that fruit quality was affected by Captan, which reduced sugar and total acid content of the

fruit but he did not report on fruit shape. Brown (1965) found that apple fruit set was greatly reduced when flowers were pollinated by hand with Captan sprayed pollen. Eaton and Chen (1969a) and Burth and Ramson (1974) showed that spraying anthers before dehiscence caused less damage to pollen than spraying dehisced anthers. Kaspars (1965) reported that sprays of Captan applied three or four times during bloom resulted in 36.3% and 64.9% reduction, respectively, in fruit set and yield of cranberries. Lockhart (1967) found that lowbush blueberries in plots treated with fungicides, were not significantly different from those in control plots with respect to the seed counts.

Bennet (1968) showed that application of fungicidal sprays to strawberry flowers increased the proportion of misshapen fruit, which subsequently reduced total commercial yield and the effect varied with the cultivar. Yet, Courly (1968) has pointed out that the beneficial effect of disease control by fungicides must be greater than any reduction in yield due to the phytotoxicity.

Captan has been shown to inhibit germination of strawberry or cranberry pollen in vitro when sprayed on or included in agar germination media (Shaw et al., 1966; Eaton and Chen, 1969a) or when sprayed on strawberry anthers after dehiscence (Eaton and Chen, 1969a). It has also been reported that strawberry pollen germination is slightly affected by Captan when sprayed on the undehisced anthers (Eaton and Chen, 1969a). In a second paper (1969b) Eaton and Chen also reported

that Captan decreased strawberry achene set and berry development and increased the proportion of misshapen fruit in the greenhouse, when sprayed after anther dehiscence. Achene set was not affected when spraying was done one day after pollination. They concluded that Captan acts directly upon germination and not upon stigma receptivity nor pollen tube growth in the style nor fertilization. Molnar (1972) reported that several fungicides used on apples (excluding Captan), which decreased pollen germination in vitro, had no effect on yield in the field. In 1973, Yor-Ganchena reported that fungicides increased the intensity of the respiration process and the vitamin-C content of strawberry. They also enhanced Polyphenoloxidase activity in cvs. "Senga Sengana" and "Souvenir". Legge and Williams (1975) confirmed that application of fungicides during blossom periods, when natural pollen transfer conditions are adverse, may affect subsequent fruit set of apples.

Generally, it has been demonstrated that most fungicides inhibit pollen germination in vitro when added to media (Rich, 1957; Gartel, 1961; Braun and Schonbeck, 1963; Cristoferi et al., 1966; Shawa et al., 1966; Eaton and Chen, 1969; Dancs and Kiss, 1970; Gentile and Vaughan et al., 1973; Church and Williams, 1977; Burth and Ramson, 1974; Bristow, 1981; Bristow and Shawa, 1981; Shawa, 1981).

Ramina (1974) recorded slight stimulation of apple pollen germination by Thiophanate-methyl and Benomyl at low pollen densities.

Others (Eaton,1961; Lockhart,1967; Eaton and Chen,1969b; Gentile and Gallagher,1972) have noted reduced germination of pollen of sweet cherries, petunias, blueberries and strawberries when the surface of the germination media had been treated with fungicides. Some workers reported that certain fungicides could reduce fruit set of apples in the field (Macdaniels and Burrell,1934; Macdaniels and Hildebrand,1940; Danoho,1964; Cristoferi *et al.*,1966; Chollet,1970). Others found variability in fruit set and pollen germination of apples from year to year (Dancs and Kiss,1970; Burth and Ramson,1974). Still others found no effect on the fruit set of apples (Schmidt,1956; Rich,1957; Kaspar,1965; Leibster,1965). Cristoferi *et al.* (1966), Dancs and Kiss (1970), Chollet (1970) and Burth and Ramson (1974) reported no effect on the final crop of apple trees. However, Shawa *et al.* (1966) reported a crop reduction of "McFarlin" cranberries caused by application of fungicides at flowering time.

In 1977, Church and Williams found that fungicide application one day before or after pollination had no effect on apple pollen germination. In 1981, Bristow and Shawa reported that among the fungicides they tested, Captan, Captafol and Mancozeb were the most toxic to cranberry pollen *in vitro*, completely inhibiting germination at 10 ug ml^{-1} . (Triforine-F and Triforine-WP did not prevent germination even at $10,000 \text{ ug ml}^{-1}$). They also found that none of the fungicides tested in the field reduced fruit set as measured by the number of

berries per upright of stem.

Jefferies, Atwood and Williams (1980) tested the effects of Carbendazin and Triforine on pollen-tube growth and cropping of goseberry cv. Careless. They used each fungicide at concentrations up to two times the strength recommended by the manufacturers, yet they observed no toxicity symptoms and no effects on fruit set, berry weight or seed number.

Church (1980) demonstrated that a wettable sulphur formulation sometimes increased, while Dodine decreased fruit set of apple trees, cv. Cox's Orange Pippin, when applied to the flower cluster.

Bristow (1981) showed that pollen grains of 13 cvs. of highbush blueberries (Vaccinium corymbosum L.), germinated on 9% sucrose agar, were inhibited when the media contained 10-50 ug ml^{-1} of Triforine. He also demonstrated that Triforine applied directly to the stigma at a concentration of 3.488 ug ml^{-1} prevented pollen germination. In addition, he found that pollen from the sprayed flowers germinated readily on sucrose agar and that fruit set and berry development were not affected by fungicide application in the field. He reported that blueberry flowers remained receptive to pollen for up to 8 days and concluded that this was why fruit set was not reduced when Triforine was applied during bloom even though it was toxic to blueberry pollen.

Shawa (1981) reported that Dithane M-45, Difolatan-F, Triforine-EC, Triforine-F, and Triforine-WP not only completely

inhibited pollen germination when applied on the surface of agar plates, but that percent fruit set was significantly reduced when fungicides were sprayed, during the blossoming period, in the field.

Recently, Howard and Albregts (1982) evaluated several different fungicides for control of strawberry fruit rot. None of treatments they used caused visible phytotoxicity (misshapen fruit) in the field. They found that none of the treatments significantly increased marketable fruit yield above that in the control.

MacSwan and Gashairi (1982), at Oregon State University, tested several fungicides for Botrytis control on strawberries. The fungicides used were Ronilon 50WP, Ronilon 50Wp, BAS 436F 50WP, BAS 436F 50WP, BAS 436F 50WP, Captan 50WP, Dyrene 50WP, Benlate 50WP, Dyrene 50WP and a control. They reported that among the fungicides tested the best control of fruit rot at the first picking was provided by Ronilon 50WP and BAS 436F 50 WP. For the whole season the best control of fruit rot was provided by BAS 436F and by Ronilon 50Wp. The largest berries were produced in plots sprayed with Ronilon 50WP 16oz. Furthermore, they concluded that none of the treatments reduced berry size. In another trial, to check the efficacy of fungicides for control of fruit rot, Moore and Bordelon (1982) observed that Captan 50WP and Benlate 50WP at different rates, together or alone, significantly reduced grey mould at harvest without phytotoxicity.

Recently Church, Cooke et al. (1983) tested the toxicity of

fungicides to apple pollen in vitro and in vivo. Their results indicated a good assessment of potential damage to pollen. Later, Church, Morgan, Cooke and Williams (1983) discussed the effects of spray volume in terms of the toxicity of Captan and Dinocap on apples in the orchard. In this paper, they indicated that a reduction of spray volume in apple orchards is likely to minimize reduction in fruit set. Further, Church and Williams (1983) tested the effects of pre-blossom fungicide spraying on the ability of Cox's Orange Pippin apple flowers to produce fruit. They concluded that most fungicides had no effect on subsequent flower opening or fruit set. However, in experiments over several years, sprays of sulphur and Dodine sometimes affected the ability of the flowers to produce fruit. Sulphur sometimes increased this ability whereas Dodine sometimes decreased it.

In view of the foregoing reports on the effects of fungicides on pollen germination and fruit set of crops other than strawberry, it was decided to carry out a similar investigation on the effect of fungicides on the strawberry crop.

These studies were conducted in order to determine whether fungicides reduce yield, stigma receptivity and pollen germination when they are used during bloom, according to Quebec recommendations, to control B. cinerea.

3. MATERIALS AND METHODS

The following studies were conducted between April 1981 and August 1982, at Macdonald College of McGill university, Ste Anne de Bellevue, Quebec, Canada.

Three strawberry cultivars were used and two fungicides applied, at various rates, in vivo and to pollen in vitro. In the field, two application times were used and in one experiment cross pollination by insects was prevented by covering the plants with fine mesh net cages.

3.1. Experiment 1. To Determine the Effects of Fungicides on Berry Quality and Yield

3.1.1. Objective of the Experiment

This study was undertaken to determine:

- 1- If Captan and Easout, applied during the blossoming period, would reduce the yield and quality of berries,
- 2- If the three cultivars (Bounty, Sparkle & Redcoat) differed in their response to fungicide application,
- 3- If the adverse effects of fungicides were dependent on when, during the flowering period, they were applied and
- 4- If the adverse effects of the fungicides might be overcome by cross pollination by insects.

3.1.2. The Cultivars

3.1.2.1. Redcoat

This is a midseason strawberry, recommended for general planting in Quebec. It is the product of a cross between Sparkle and Valentine which was introduced in 1957 by the Ontario Research Station, Ottawa, Ontario. Redcoat is the main cultivar in Eastern Canada (Craig, 1982).

The plant is vigorous and hardy and develops extensive runners (Craig, 1982). Redcoat is the highest yielding cultivar at Vineland, Ontario, producing medium, firm fruit of good size, whose later fruits (secondary, tertiary, etc.) maintain their size well (Darrow, 1966; Rickeston et al., not dated; Craig, 1982). It is very attractive after storage, having medium sized red fruit with good shipping quality. However, it is very susceptible to B. cinerea fruit rot and is also susceptible to Verticillium wilt (Craig, 1982).

3.1.2.2. Sparkle

(Paymaster) is a midseason strawberry, recommended for limited planting in Quebec. Sparkle is the product of a cross between Fairfax and Aberdeen which was introduced in 1942 by the N.J. Agricultural Experimental Station, New Brunswick, N.J.. The plant is vigorous and develops extensive runners. It is also productive and has attractive fruit, medium light to medium dark red colour, soft and with good

flavour. However, Sparkle is susceptible to Verticillium wilt, leaf scorch and leaf spot (Darrow, 1966; Ricketson et al, not dated; Craig, 1982).

3.1.2.3. Bounty

This is a late-season strawberry recommended for general planting by Agriculture Canada. It was introduced in 1972 from the Agriculture Canada Research Station, Kentville, N.S. Bounty is a cross between Jerseybelle and Sengasengana. The plant is vigorous and develops runners well. It is also very productive. The fruit has medium to medium dark red skin, almost dull and seedy in appearance, is moderately firm and has good flavour. The first fruits are very large, but later fruits are small to medium in size. Bounty's foliage is resistant to leafspot but is susceptible to Verticillium wilt and slightly susceptible to leaf scorch. It is somewhat resistant to Botrytis fruit rot (Ricketson et al, not dated; Craig, 1982).

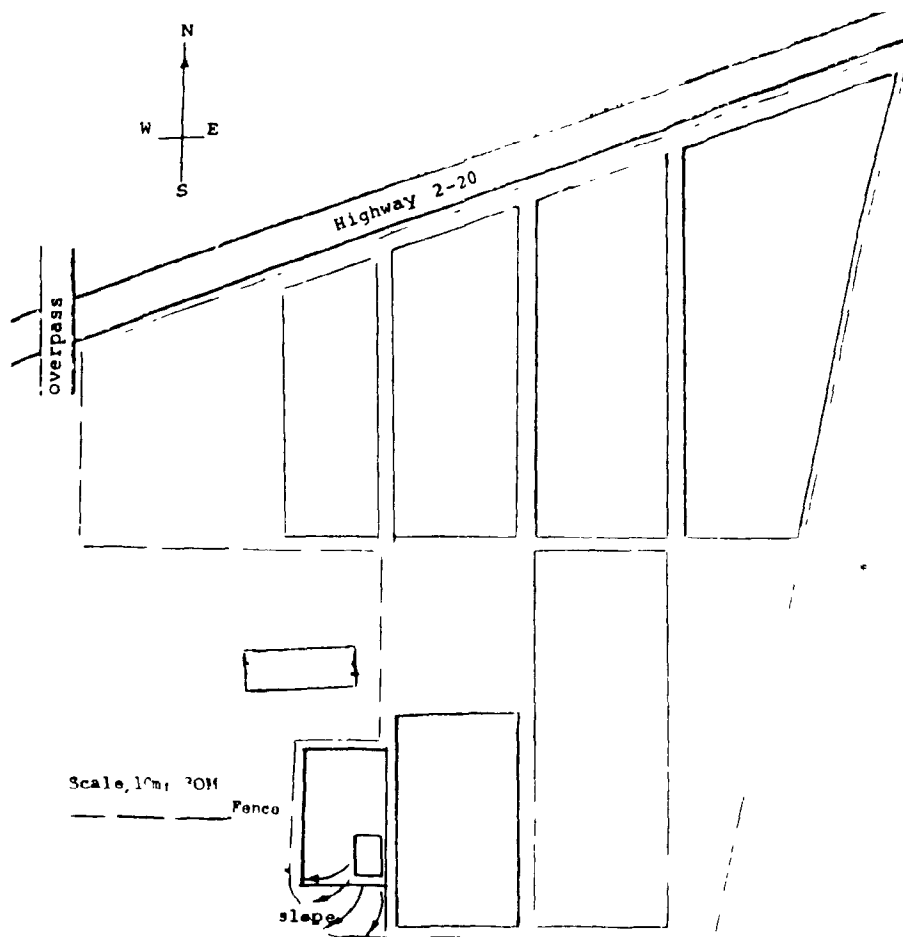
Certified plants of these three cultivars were obtained from Luc Lareault, Lavaltrie. All the plants were in good condition. However, the Redcoat and Sparkle plants looked a little smaller and had fewer leaves than the Bounty plants at the time of planting.

3.1.3. Field Preparations and Planting

The field used for this experiment was 30 X 18m, located on a hill with a slight slope toward the southwest (Fig. 6). The field was plowed and 675kg ha^{-1} of fertilizer (20-20-20) added before rototilling (disking). Despite plowing, however, at the time of transplanting block 1 had compacted soil compared with the other blocks.

Planting took place on May 7th 1981 and plants were set 45cm apart in each row and 80cm apart between rows, using a bulb planter. The roots of plants were not cut back at the time of transplanting in order to allow better vegetative and reproductive growth in the first year. All dead and broken leaves were removed, and the three youngest leaves were retained. One half cup of starter solution (10-52-10), 5mg l^{-1} was applied to each plant at the time of transplanting to promote rooting.

A total of 1440 rooted runners (280 of each cultivar) were planted. Before treatment applications, live rooted plants were exchanged for any dead ones. It is commercial practice to deblossom plants in their first year to encourage the development of runners, however, for this experiment plants were allowed to bear flowers the first year and these were used in the field trial in 1981. All plants produced at least one cyme; any extra inflorescences were removed prior to treatment applications.



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Figure 6. Location of the field experiment (1981 & 1982), Horticulture facility, Macdonald College of McGill University, Ste. Anne de Bellevue.

3.1.4. Caging

To prevent pollen transfer from untreated to treated flowers, by pollinating insects, 72 individual cages were made. These cages were constructed using three wires and 3 m of mesh net each (Fig. 7).

The height of each cage was approximately 30 cm and completely covered five plants over an area of 45 X 225cm (one experimental unit).

Light intensity was measured, at the level of the plants, using a Weston illumination meter (model 756 with quartz filter). The intensity was measured for both caged and uncaged plants in a total of ten different locations. This was done to determine the per-cent reduction of light caused by the mesh net. The average light intensity was 9730 F.C. outside of the cages and 8870 F.C. inside cages. Thus the cages caused a reduction of light intensity of less than 9% (Fig. 8).

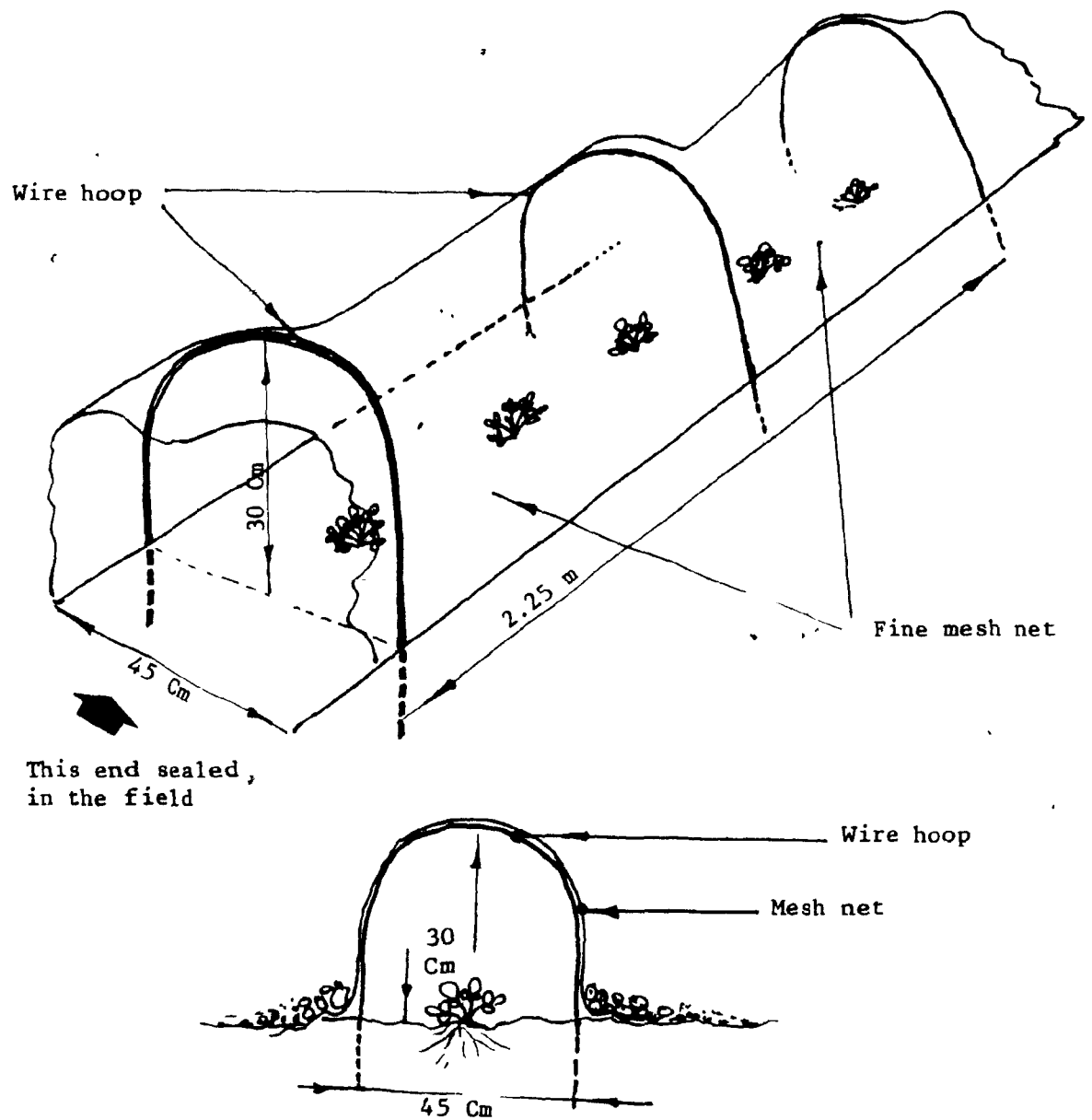


Figure 7. A schematic view of cage using fine mesh net and three wires to prevent the entry of pollinating insects.

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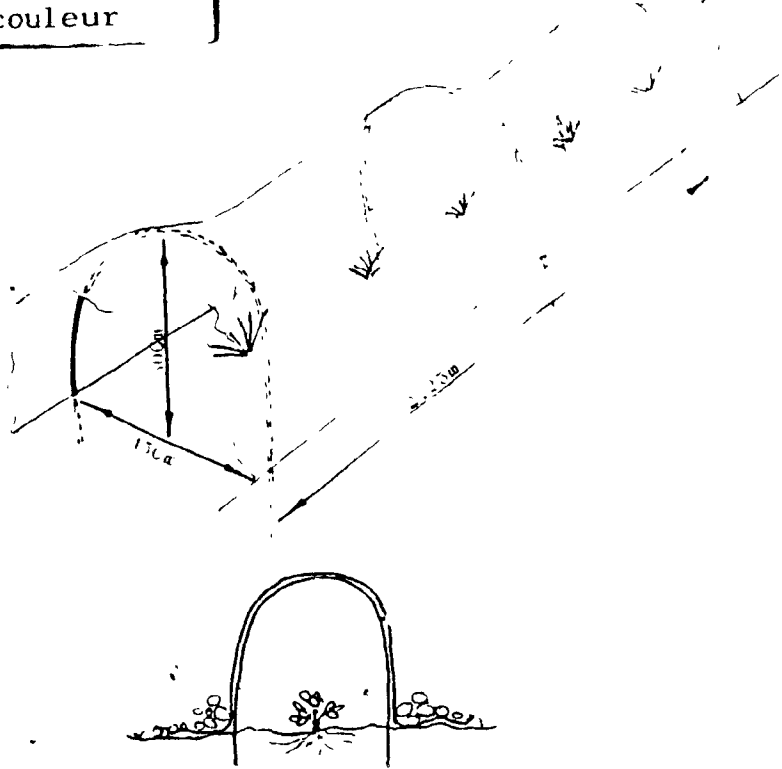


Figure 8. View of fine mesh net cages constructed to prevent the entry of pollinating insects.

3.1.5. Applications of Fungicides

Two fungicides, Captan and Easout were applied. Captan 50% WP was applied at a rate of 2g l^{-1} and Easout 70% WP at a rate of 1g l^{-1} to the plants using a 22 litre packspray until run-off occurred. Early applications were made when the primary flowers were open and mid applications were made when the flowers were in late full bloom. Fungicide applications started on June 1 and finished on June 11, depending on the particular cultivar as follows:

<u>Cultivar</u>	<u>Stage of flowering</u>	<u>Date of fungicide application</u>
Redcoat	Early bloom	June,01,1981
Redcoat	Full bloom	June,05,1981
Sparkle	Early bloom	June,05,1981
Sparkle	Full bloom	June,08,1981
Bounty	Early bloom	June,08,1981
Bounty	Full bloom	June,11,1981

3.1.6. Application of Insecticides

To prevent damage by spittle bugs (Fig. 9) and two species of moths (Fig. 10), the larva of which caused damage to strawberry leaves in the field (Fig. 11), Malathion 25% WP was used at a rate of 1.2g l^{-1} . The spray was applied on June-15 to all the experimental units after the flowering period.



Figure 9. Spittle bugs observed on strawberry plants in 1981 before insecticide application.

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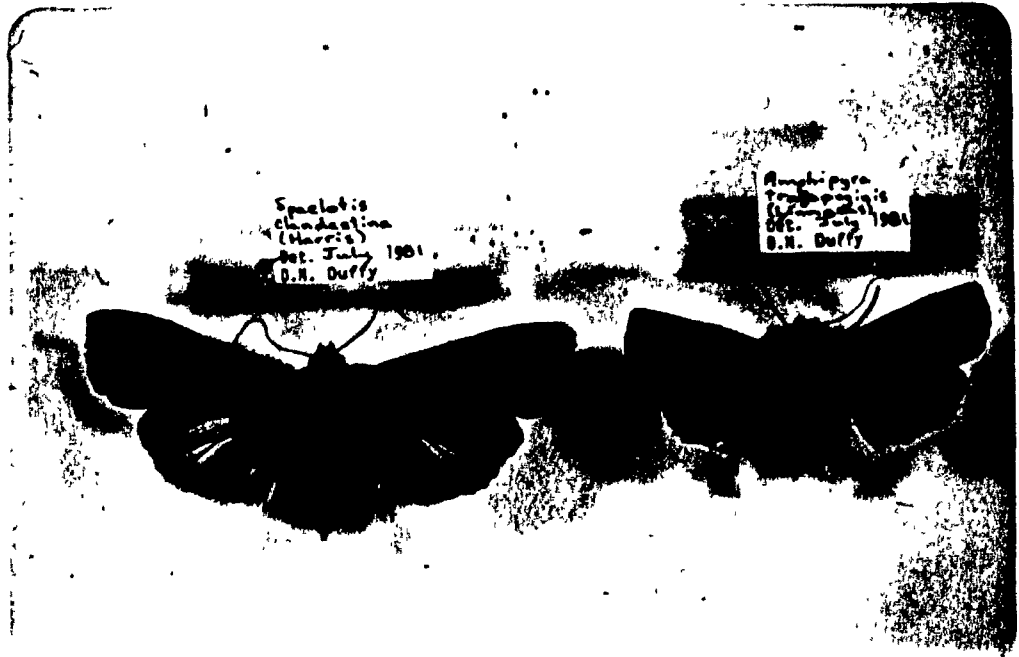


Figure 10. Two moths (Spaelotis and Ampipyra) hatched from larvae found on strawberry plants in the field in 1981.



Figure 11. Larva on a strawberry leaf.

3.1.7. Soil Testing

Nine soil samples from three different parts of the field were collected. Each sample contained a mixture of soil taken from 10cm and 20cm depths. A Lamotte combination soil testing outfit (model STH) was used and the results are presented in appendix. 3, Table.1.

The soil was not kept frozen between collecting and testing, therefore, results of the nitrate and nitrite nitrogen analyses are not accurate. The pH of the soil was found to be between 6.6 and 6.2 and there was an adequate amount of fertilizer. The textural class of the soil was considered to be clay loam (very close to sandy clay loam). See appendix. 3. "Procedure for particle size analysis".

3.1.8. Field Management

Weeds were removed with a cultivator and Dutch hoe. In addition, on July, 3 1981, Tenoran 50% WP (at a rate of 4.0 kg h^{-1}) was used to keep the field free from broad leaf weeds. Unfortunately the cultivar Bounty is very sensitive to Tenoran and as a result leaf damage was so severe that 50% of the leaves were killed.

3.1.9. Factors Used in this Experiment

The factors in this experiment were:

- 1) Three cultivars (Redcoat, Sparkle and Bounty),
- 2) Two fungicides plus a water control (Captan + tap water, Easout + tap water and tap water),
- 3) Two times of fungicide applications (early and late full bloom) and
- 4) Two types of pollination control (caged and uncaged).

3.1.10. Experimental Design and Procedure

Split plots were used to assign the above factors in a randomized complete block design with four blocks.

The types of pollination were randomized within the blocks in each main plot unit. The three cultivars were randomized in each sub-plot unit in each of the main plot units. Two different times of fungicide application were randomized to the sub-sub-plot units in each sub-plot unit and two fungicides + control were randomized to the sub-sub-sub-plot units in each of the sub-sub-plot units. See appendix 4., Fig. 1.

Each sub-sub-sub-plot unit contained five plants and was separated from other experimental units by two plants at one end and three at

the other.

To reduce loss of fruit by bird damage, the fruit was harvested when only 75% of the berry surface had turned red. Flower numbers, fruit numbers, total fruit weight per plant, per-cent fruit set, fruit disease, fruit shape and mean fruit weight were measured separately for 720 strawberry plants. Nine fruits from each plot (five plants) were randomly selected at three different harvest times (first harvest, mid harvest and final harvest) and kept frozen until the end of the experiment. This was done to determine the number of abortive achenes, sound achenes, total achenes and per-cent of sound achenes per berry (Fig. 12).

The total number of achenes were counted on 1296 berries. Each berry was cut in half and then each half was pressed between two pieces of glass. A cell counter was used to count the number of achenes on each berry.

The data from 36 experimental units in each block were analysed by analysis of variance. Comparisons were made using Duncan's new multiple range test and the least significant difference was determined, at a 0.05 level of significance. Fruit shape and disease were ranked from 0 to 10. In ranking fruit shape, 0 was awarded to totally malformed fruit and 10 to perfectly shaped fruit. In ranking fruit disease, 0 was awarded to totally infected fruit and 10 to disease free fruit.

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aborted achenes

sound achenes

style

stigma



Figure 12. Developing fruit of the cv. Bounty, showing both fertilized and aborted achenes on the receptacle.

All the data which was not normally distributed (i.e. number of achenes and ranking for shape and disease) were transformed before analysis of variance, using Log or square root transformations as required (Steel and Torrie, 1980).

3.2. Experiment 2. To Determine the Effects of Fungicides on Pollen Viability and Stigma Receptivity

3.2.1. Objectives of the Experiment

The results of experiment 1 appeared to contradict the results of other workers (Eaton and Chen, 1969a; Eaton and Chen, 1969b) with respect to berry quality and yield, therefore, a second experiment was undertaken to confirm the results.

The objectives were to determine the effects of the fungicides Captan and Easout on pollen viability and stigma receptivity of three strawberry cultivars (Sparkle, Redcoat and Bounty). In addition, to find out if the fungicides had any effects on berry quality or yield (or both) in a closed chamber (greenhouse).

3.2.2. Pollen germination test

For the purpose of comparison, a preliminary pollen germination test was done prior to examining the effect of fungicides on pollen germination as follows:

A random sample of anthers from the three cultivars was

examined to determine the percent pollen germination (viability). To do this, ten microscope slides were prepared as follows: One drop of a 10% sucrose solution plus pollen mixture was placed on a glass cover slip. This was then located upside down over a square wall of petroleum jelly, on a glass slide (Hanging drop method), (Bishop, 1949; Sharma and Sharma, 1972); (see appendix. 4, Fig. 2). These slides were incubated at room temperature. At the end of three hours, the percent pollen germination was recorded by counting after 100 pollen grains. The average percent germination was over 90% in vitro before treatment application.

3.2.3. Treatments

The seven treatment combinations used were broken down as follows:

- a) three were to study the effect of fungicides on pollen germination,
- b) three were to study the effect of fungicides on pistils and
- c) one was an untreated control.

To study the effect of fungicides on pollen, a mixture of pollen taken from the three cultivars was collected and air dried overnight in the laboratory. The dried pollen mixture was then divided into three groups. The first group was sprayed with Captan 50% WP (2g l^{-1}), the

second group was sprayed with Easout 70% WP (1g l^{-1}) and the third group was sprayed with tap water. The pollen was sprayed until totally wetted using the recommended commercial dilutions.

The treated pollen mixtures were left to dry (to avoid contamination of the stigma with wet fungicide remaining on the pollen). The dried pollen was applied to emasculated flowers. A different, clean camel hair brush was used for each of the three treatment combinations.

To study the effects of fungicides on pistils, the emasculated flowers were divided into three groups. The first group was sprayed with Captan 50% WP (2g l^{-1}), the second group was sprayed with Easout 70% WP (1g l^{-1}) and the third group was sprayed with tap water. These flowers were sprayed until run-off, then allowed to dry completely before pollination.

Previous to this, a mixture of pollen taken from the three cultivars was collected and air dried overnight in the laboratory. This pollen was left untreated and applied to the dried treated flowers using a different camel hair brush for each of the three treatment combinations.

For the untreated control, the emasculated flowers received neither fungicides nor pollen.

3.2.4. Experimental Design and Procedure

This experiment commenced on October 13th, 1981, in the greenhouse. The strawberry plants used were daughter plants, of uniform size, taken from the three certified cultivars (Redcoat, Sparkle, Bounty) used in the previous experiment (summer 1981). Of the total of 168 plants used, 56 were selected from each cultivar. These were grown in a mixture of loam, peat, sand and vermiculite (2:1:1:1 volume), in six inch, shallow, plastic pots.

To promote root initiation, 5 mg l⁻¹ fertilizer (10-52-10) was added to each pot. Beginning two weeks after transplanting, the plants were fertilized weekly (with mixture of 13-20-20 1g l⁻¹ prepared by the author) at a rate of one cup per pot. This was done to maintain optimal growth. In addition, Sodium tetraborate and Calcium nitrate were applied five times during the experiment to prevent calcium and boron deficiency.

The plants were exposed to 16 hours of light each day. To achieve this, the natural daylight was supplemented with a combination of high pressure sodium and incandescent lamps. The temperature in the greenhouse was maintained between 20-23°C during the 16 hour "day" and 13-18°C during the night.

A randomized complete block design was used involving 168 pots.

Each block contained 42 pots. Twenty one treatment combinations were used; one treatment combination for each experimental unit. Each of these experimental units consisted of two pots; each containing one strawberry plant.

For the purpose of this experiment, only the two secondary flowers on each plant were retained and each of these was emasculated before fully opened (Fig. 13).

Berry weight per plant, fruit disease, fruit shape, and the number of aborted and sound achenes were all recorded. Then, the average of two sampling units in each experimental unit was calculated. These averages were used for comparison among the treatment combinations, for the above variables, using the general linear model procedure.

For convenience in ranking fruit shape, a scale between 0 to 4 was used. This was done since in the previous experiment the scale of 0 to 10 was found to be too large. Zero was awarded to malformed fruit and 4 for perfect fruit shape.

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Figure 13. Stage of development at which flowers were emasculated, before treatment application in the greenhouse experiment.

3.3. Experiment 3. To Determine the Effects of Fungicide Concentration on Pollen Germination and Length of Pollen Tubes

3.3.1. Objective of the experiment

The results of experiment 2. indicated that fungicides could have a significant adverse effect on berry quality and yield. This experiment was conducted to determine the effect of fungicide concentration. The experiment was carried out in vitro using fungicides at different concentrations in the pollen germination medium.

3.3.2. Factors used in this experiment

Three strawberry cultivars (Sparkle, Redcoat and Bounty) were used and two fungicides (Captan 50% WP and Easout 70% WP) at a range of concentrations:

- 1) Captan 50% WP at 2000ppm incorporated into the 10% sucrose solution,
- 2) Captan 50% WP at 1000ppm incorporated into the 10% sucrose solution,
- 3) Captan 50% WP at 500ppm incorporated into the 10% sucrose

solution,

- 4) Easout 70% WP at 2000ppm incorporated into the 10% sucrose solution,
- 5) Captan 70% WP at 1000ppm incorporated into the 10% sucrose solution,
- 6) Captan 70% WP at 500ppm incorporated into the 10% sucrose solution, and
- 7) 10% sucrose solution without any fungicide (control).

3.3.3. Experimental Design and Procedure

Anthers from the three cultivars were collected separately in the evening and allowed to dehisce in the laboratory. The fungicides, at the specified range of concentrations, were then incorporated into the 'pollen tube growth-medium' (a 10% sucrose solution). A small amount of pollen was transferred into the liquid by means of a small brush. One drop of each of the 'liquid + pollen' mixtures was put onto a glass cover slip and located upside down over a square well of petroleum jelly on a glass microscope slide (hanging drop method), (Bishop, 1949; Sharma & Sharma, 1972); (see appendix. 4, Fig. 2).

These slides were incubated at room temperature for different lengths of time (three, six, nine and twenty-four hours). Then the percent pollen germination and the length of pollen tube were recorded

for the different intervals of time. For the purpose of calculating the percent pollen germination, 100 pollen grains were counted on each slide.

The laboratory experiment was conducted as a split-plot in time, in a completely randomized design, with three replicates. A total of 252 observations were made of 84 treatment combinations (three cultivars X seven fungicide concentrations X four different examination times).

3.4. Experiment 4. To Determine the Effects of Fungicides on Berry Quality and Yield and the Role of Insect Pollinators

3.4.1. Objective of the Experiment

This experiment was undertaken to confirm the results obtained in experiment 1. The objectives were to determine:

- 1) if the fungicides, Captan and Easout, applied during the blossoming period would reduce yield and increase the number of malformed berries and
- 2) if the three cultivars (Sparkle, Redcoat and Bounty) differed in their response to fungicide application.

In addition observations were made of the number of insects that visited the strawberry flowers, both during and after fungicide application

3.4.2. Factors used in this experiment

The factors in this experiment were:

- 1) three cultivars (Sparkle, Redcoat and Bounty) and
- 2) two fungicides plus a water control (Captan + tap water, Easout + tap water and tap water).

3.4.3. Experimental Design and Procedure

Overall, 180 plants were examined. These plants were protected by straw mulch during the winter of 1981-1982. As application timing and caging had already been tested in experiment 1, these factors were omitted. Two fungicides plus a water control in split-plots with four blocks were used. Each block was divided into three main plots and the cultivars were randomized in each block. Each main plot was divided into three sub-plots and the fungicides and the water control were applied to each sub-plot randomly. The fungicides were applied at the commercially recommended rates (2g l^{-1} Captan 50% WP & 1g l^{-1} Easout 70% WP). Fruit number, weight, shape, disease, the total number of achenes, abortive and sound achenes were recorded for each of the individual plant's fruits. Analysis of variance was performed on the mean of the four plants in each experimental unit. Insects visiting the flowers were observed.

4. RESULTS

4.1 Experiment 1. The Effects of Fungicides on Fruit Quality and Yield

Analysis of variance showed there were no significant interactions between caging, cultivars, time of fungicide application or type of fungicides used. See appendix. 1, tables 1 to 7. Due to the independent effects of factors on the variables, each dependent variable was treated separately and the results were as follows:

4.1.1. Flower and Fruit Number

There were significant differences between cultivars in terms of flower and fruit number. Bounty ranked best of the three cultivars tested (table 1). There were no differences between caged or uncaged plants in terms of the number of flowers and fruit (table 1).

Table 1. Flower and fruit numbers of three strawberry cultivars grown under caged and uncaged conditions.

factors	cultivars			caging	
	Redcoat	Sparkle	Bounty	caged	uncaged
flower number	8.16b	7.97b	9.85a	8.99	8.32
fruit number	6.08c	6.59b	7.93a	7.12	6.61

Duncan's new multiple range test.

Cv means with the same letter are not significantly different at 0.05 level of significance.

Each figure is the mean of 48 observations.

As expected, the type of fungicide (Captan or Easout vs. water) had no significant effect on the number of flowers, regardless of whether they were applied at early or late full bloom (table 2), nor did they influence fruit number.

4.1.2. Percent Fruit Set and Fruit Weight

There were highly significant differences between cultivars in terms of percent fruit set. Bounty and Sparkle set the most fruit (table 3). $\log_{10}$ conversions of the percent fruit set were analysed and similar results were obtained (table 3). Fungicides and the time of application had no effect on percent fruit set (Fig. 14) or fruit weight (Fig. 15), (table 4).

Table 2. Flower and fruit numbers following fungicide application at early or late bloom.

factors	fungicide			time of application	
	Captan	Easout	water	early	late
flower number	8.37	8.88	8.73	8.69	8.63
fruit number	6.55	7.00	7.04	6.95	6.77

Captan 50% WP, 2g l⁻¹, and Easout 70% WP, 1g l⁻¹

Table 3. Percent fruit set and fruit weight of three strawberry cultivars grown under caged and uncaged conditions.

factors	cultivars			caging	
	Redcoat	Sparkle	Bounty	caged	uncaged
percent fruit set	75.66b	83.44a	80.43a	79.33	80.35
Log percent fruit set *	1.85b	1.90a	1.89a	1.87	1.89
fruit weight	37.15c	41.13b	58.70a	47.28	44.04

Duncan's new multiple range test.
 Cv means with the same letter are not significantly different, $p = 0.05$.

Each figure is the mean of 48 observations.

* Log_{10} conversion for analysis of variance procedure.

Table 4. Percent fruit set and fruit weight of strawberries following fungicide application at early or late bloom.

factors	fungicides			time of application	
	Captan	Easout	water	early	late
percent fruit set	79.35	78.78	81.41	82.18	79.50
Log percent fruit set*	1.87	1.87	1.90	1.88	1.87
fruit weight (g)	43.25	46.30	47.43	46.32	44.92

Captan 50% WP, 2g l^{-1} and Easout 70% WP, 1g l^{-1}

* Log_{10} conversion for analysis of variance procedure.

Each figure is the mean of 48 observations.

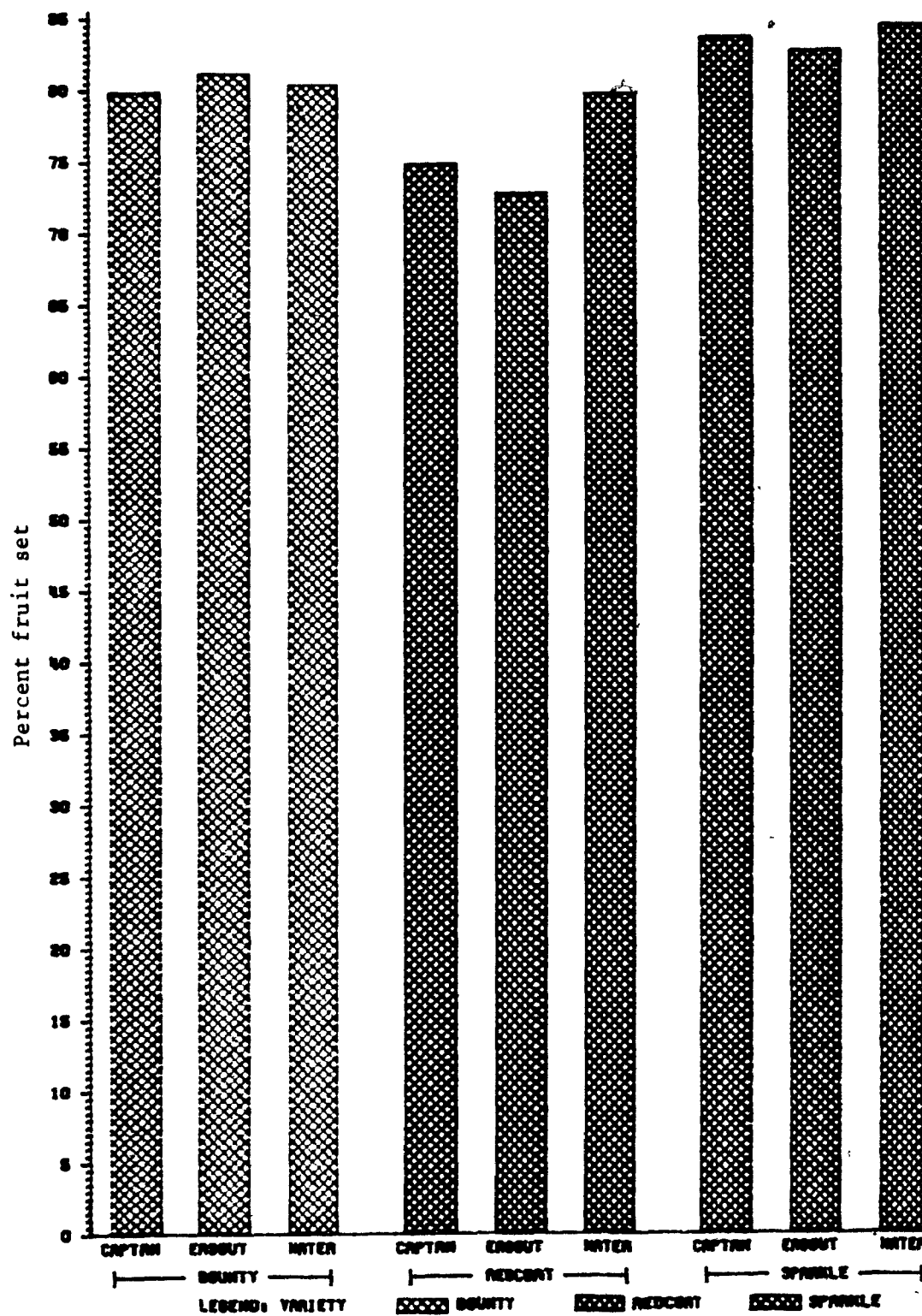


Figure 14. The effect of fungicide treatment on fruit set of three strawberry cvs. Each bar represents the mean of 16 observations.

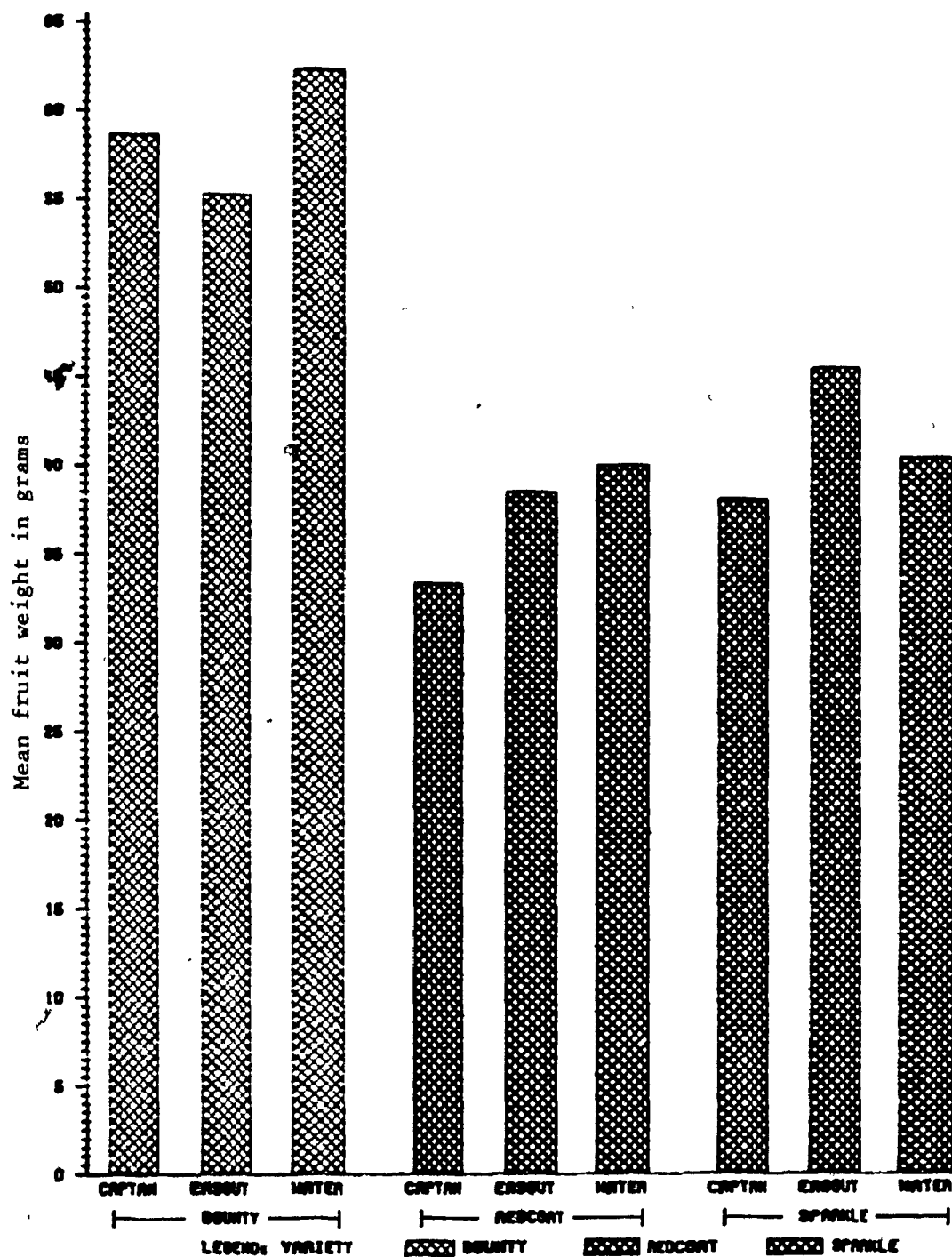


Figure 15. The effect of fungicide application on mean fruit weight (g) per plant. Each bar represents the mean of 16 observations.

4.1.3. Fruit Shape and Disease Rating

There were significant differences between cultivars in terms of fruit shape. Bounty produced the best fruit (Fig. 16) and Redcoat produced the most malformed fruit (table 5). In terms of fruit disease, there was no difference between Sparkle and Bounty (Fig. 17). Redcoat was the most susceptible to B. cinerea (table 5).

Table 5. Fruit shape and disease rating of three strawberry cultivars grown under caged and uncaged conditions.

factors	cultivars			caging	
	Redcoat	Sparkle	Bounty	caged	uncaged
fruit shape	6.3c	7.9b	8.4a	7.4	7.7
fruit disease	8.9b	9.7a	9.9a	9.3a	9.7b

Duncan's new multiple range test.

Cv means with the same letter are not significantly different, $p = 0.05$.

Each figure is the mean of 48 observations.

fruit shape: 0=malformed fruit, 10=perfect shape.

fruit disease: 0=totally infected, 10=no disease.

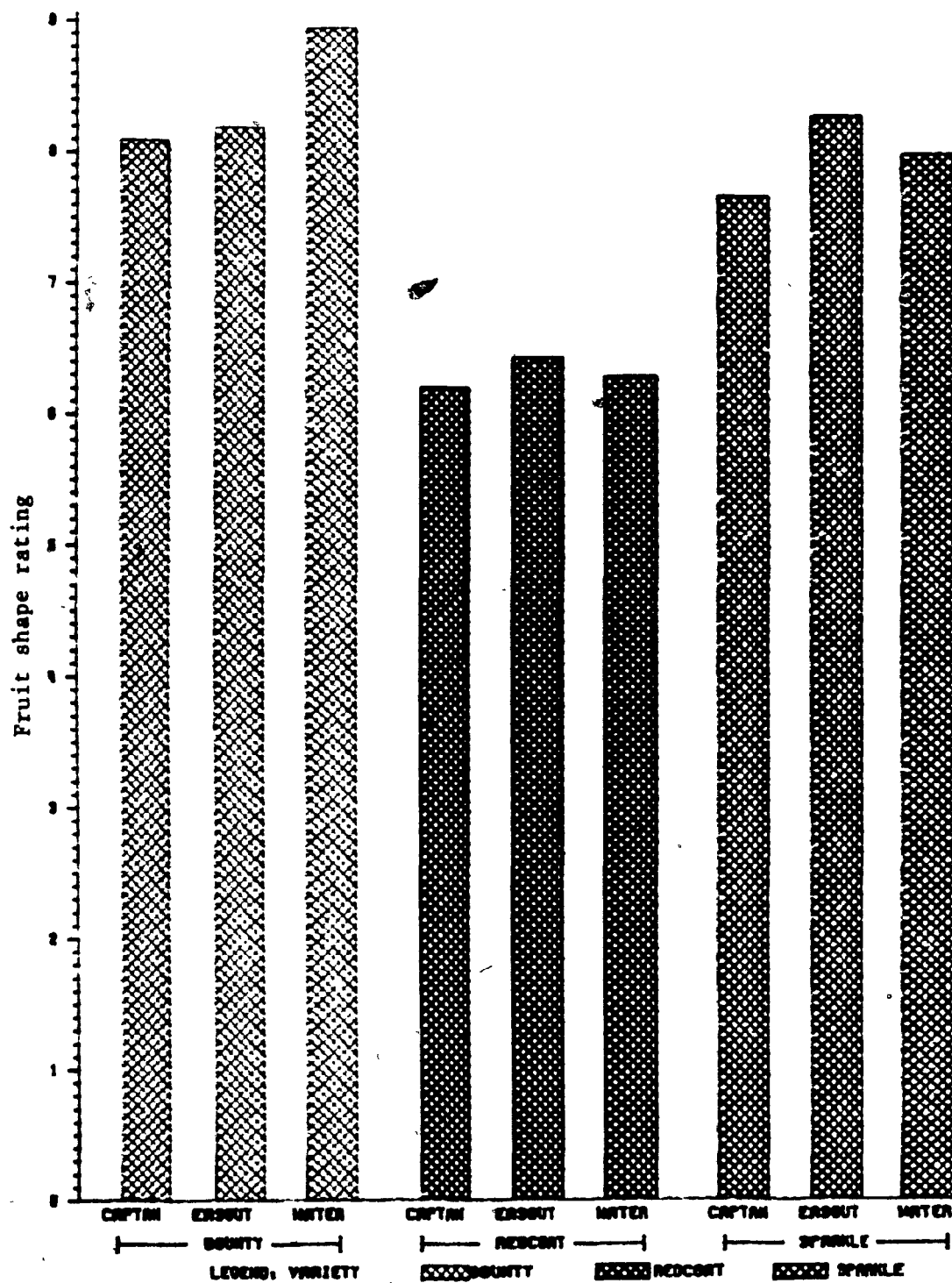


Figure 16. The effect of fungicide application on fruit shape. 0 = malformed fruit; 10 = perfect shape. Each bar represents the mean of 16 observations.

Caging did not effect fruit shape but a significant infection of the berries by B. cinerea was found among the caged strawberries (table 5). Fungicide application did not increase the number of malformed berries compared with the control (Fig. 16), (table 6). The incidence of fruit disease was not significantly different (at the 0.05 level) for Easout, Captan or water applications (Fig. 17). Nevertheless, there was a significant difference, if only at the 0.07 level (table 6). It appears that there was no significant difference between early or late fungicide application on fruit disease but the number of malformed fruits were significantly increased when fungicides were applied early in the season (first bloom), (table 6).

Table 6. Fruit shape and disease of strawberries following fungicide application at early or late bloom.

factors	fungicides			time of application	
	Captan	Easout	water	early	late
fruit shape	7.29	7.60	7.70	7.32b	7.75a
fruit disease **	9.55ab	9.60a	9.31b	9.38	9.59

** Significant at 0.07 level.

Duncan's new multiple range test.

Cv means with the same letter are not significantly different, $P=0.05$.

Captan 50% WP, $2g\ l^{-1}$ and Easout 70% WP, $1g\ l^{-1}$

fruit shape: 0=malformed fruit, 10=perfect shape.

fruit disease: 0=totally infected, 10=no disease.

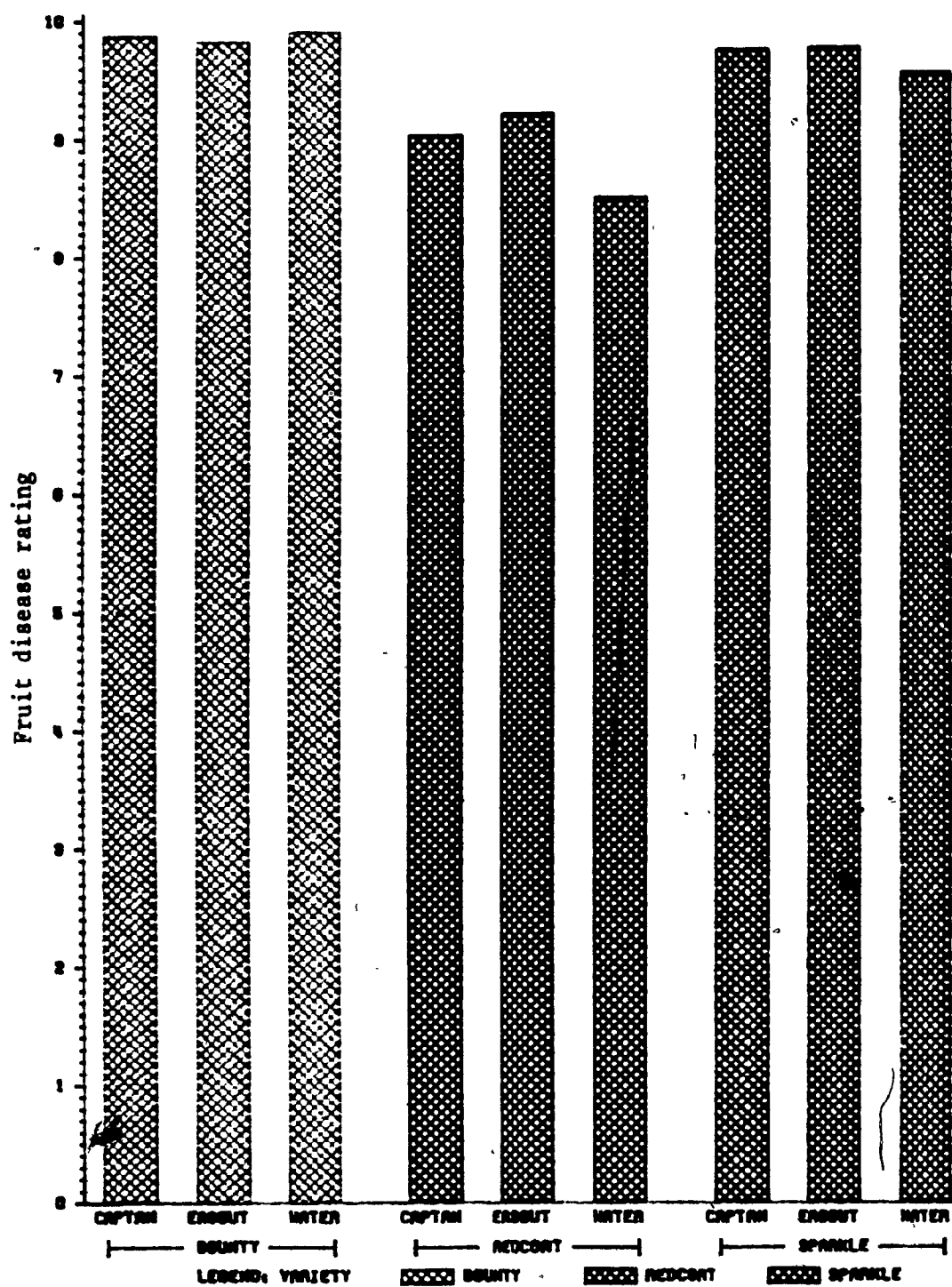


Figure 17. The effect of fungicide application on fruit disease. 0 = totally infected; 10 = no disease. Each bar represents the mean of 16 observations.

4.1.4. Number of Achenes

There were highly significant differences between cultivars in terms of the number of sound achenes (table 7). Redcoat had the most aborted achenes (table 7). Of the three cultivars tested, Bounty had the fewest aborted achenes (table 7). There was no significant difference between Sparkle and Bounty in terms of number of sound achenes and both had significantly more than Redcoat (table 7).

Table 7. The effect of caging and cultivars on achene number.

factors	cultivars			caging	
	Redcoat	Sparkle	Bounty	caged	uncaged
aborted achenes	159a	143b	140c	162a	132b
sound achenes	325b	492a	467a	409b	447a
total achenes	484c	635a	607b	571	579
sound achenes **	5.35b	5.62a	5.63a	5.48b	5.58a

Duncan's new multiple range test.
Cv means with the same letter are not significantly different,
p = 0.05.

Each figure is the mean of 48 observations.

** Log₁₀ conversion (of percent sound achenes) for analysis of variance procedure.

For statistical purposes Log_{10} transformation of sound achenes data were also tested. The results were similar to the non-transformed data (table 7). According to these results, caging led to more achenes being aborted and fewer sound achenes. Again, transformation of sound achenes data to Log_{10} percent of sound achenes did not affect the analysis (table 7). Fungicides significantly increased the number of aborted achenes and reduced the number of sound achenes (Fig. 18). Log_{10} conversion of percent sound achene data showed similar results (table 8). Early fungicide application increased the number of aborted achenes at the 0.05 level of significance and reduced the number of sound achenes (table 8).

Table 8. Achene number per berry following fungicide application at early or late bloom.

factors	fungicides			time of application	
	Captan	Easout	water	early	late
aborted achenes	160a	158a	124b	153a	141b
sound achenes	426b	416b	443a	414b	442a
total achenes	576	582	567	567	583
sound achenes **	5.47b	5.51b	5.61a	5.50b	5.56a

Duncan's new multiple range test.

Cv means with the same letter are not significantly different, $p = 0.05$.

Captan 50% WP, 2g l^{-1} and Easout 70% WP, 1g l^{-1}

** Log_{10} conversion for analysis of variance procedure.

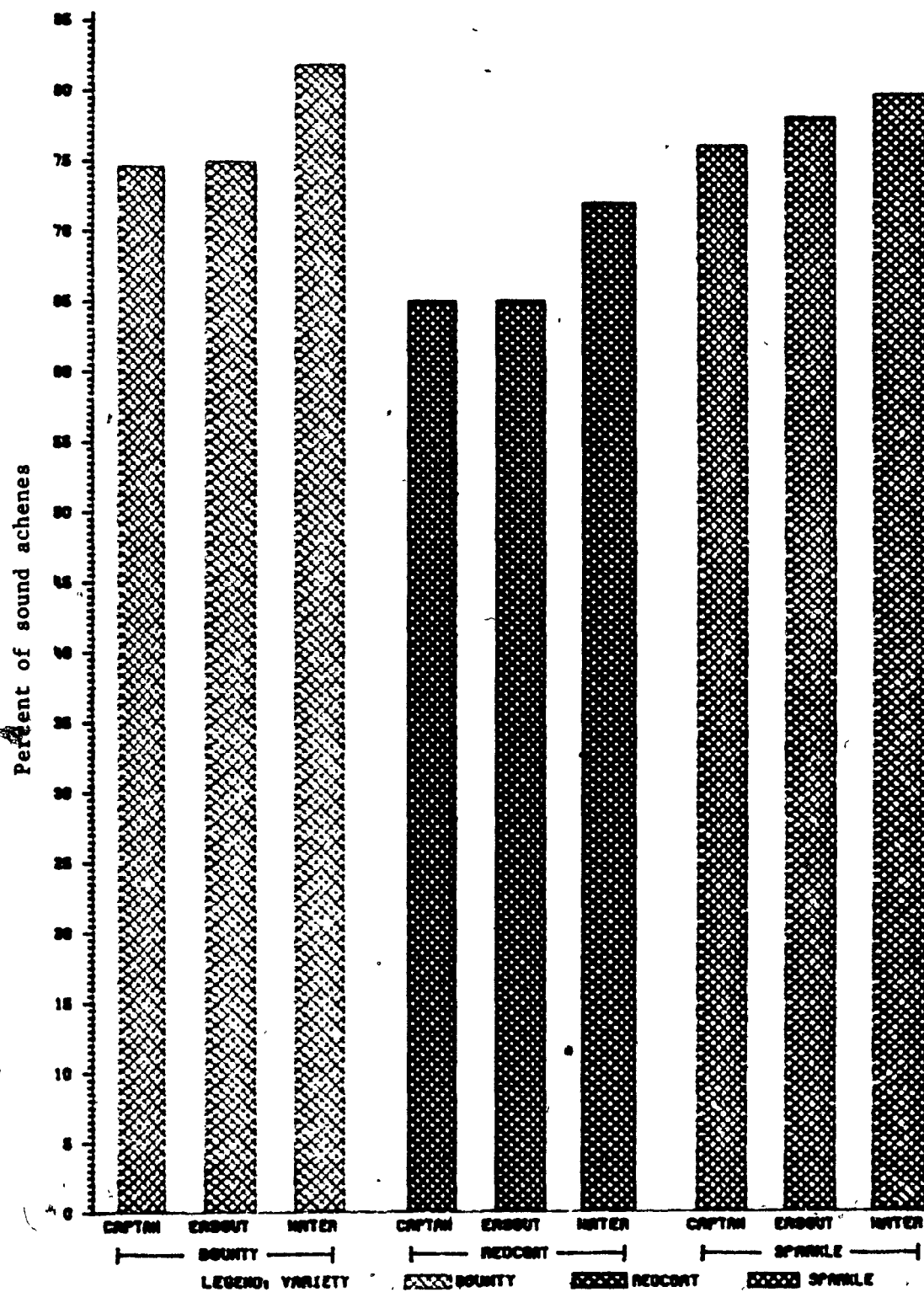


Figure 18. The effect of fungicide on percent sound achenes. Each bar represents the mean of 16 observations.

4.2. Experiment 2. Effects of Fungicides on Pollen Viability and Stigma Receptivity

Analysis of variance showed that in all cases there was a highly significant interaction between the factors tested. The effects of cultivar and fungicide were not independent in this experiment (see appendix. 1, tables 8 to 14).

4.2.1. Berry Weight

According to the results, applying fungicide to the pollen significantly reduced yield at the 0.01 level of significance. Captan had the most detrimental effect (table 9), (Fig. 19). Application of fungicides to the stigma did not significantly reduce fruit weight.

There were differences between the cultivars in response to fungicide application. The berry weights of Sparkle and Redcoat were reduced the most. Bounty ranked best of the three cultivars (table 9).

4.2.2. Berry Shape

There were no differences between berries produced after treatment of the stigma with Captan, Easout or water, or those berries produced after application of pollen treated with water. However, those produced after application of pollen treated with Captan and Easout were malformed. Captan had the most detrimental effect (table 10),

Table 9. Mean individual fruit weight (g) of three strawberry cultivars following treatment of either pollen or stigma with fungicides. (All flowers were emasculated before treatment application).

treatments	cultivars		
	Bounty	Redcoat	Sparkle
Control-unpollinated	0.24	0.21	0.30
Pollen + Captan	7.48	3.56	4.52
Pollen + Easout	7.44	4.51	9.12
Pollen + water	13.47	12.43	10.05
Stigma + Captan	11.66	8.78	8.94
Stigma + Easout	8.92	9.27	9.65
Stigma + water	10.49	10.85	10.60

LSD., (0.05) = 2.10

LSD., (0.01) = 2.84

DF=60, MS=2.2762

Each figure represents the mean of four observations.

Captan 50% WP, 2g l^{-1} and Easout 70% WP, 1g l^{-1} .

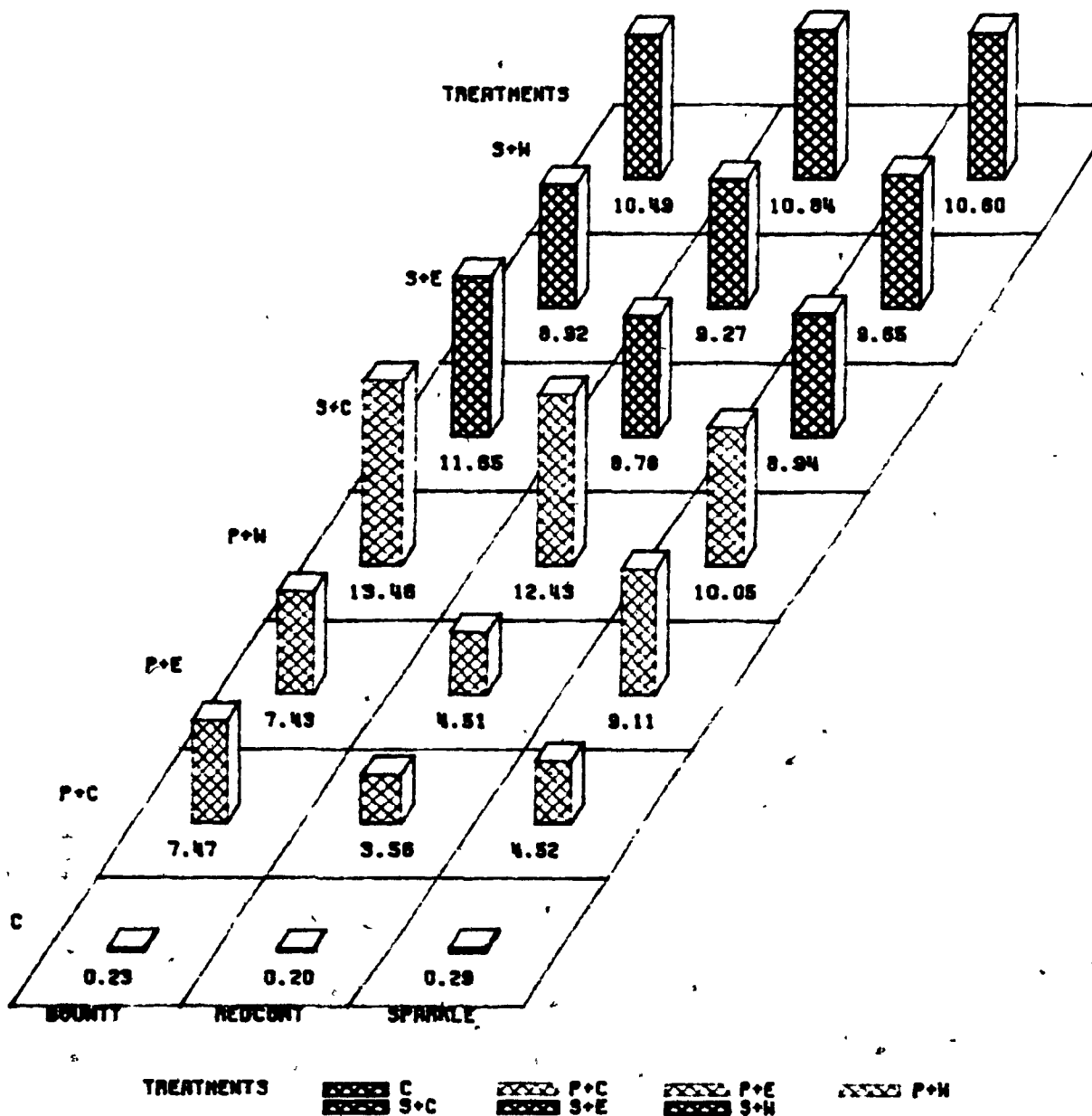


Figure 19. Mean individual fruit weight (g) of three strawberry cvs. following treatment of either stigma or pollen with fungicides. Each bar represents the mean of eight observations.
 C = control P+C = pollen + Captan P+E = pollen + Easout
 S+C = stigma + Captan S+E = stigma + Easout S+W = stigma + water

Table 10. Berry shape rating of three strawberry cultivars following treatment of either pollen or stigma with fungicides. (All flowers were emasculated before application).

Treatments	cultivars		
	Bounty	Redcoat	Sparkle
Control-unpollinated	0.00	0.00	0.00
Pollen + Captan	3.25	1.00	1.63
Pollen + Easout	3.50	1.75	3.38
Pollen + water	3.88	3.88	4.00
Stigma + Captan	4.00	3.88	4.00
Stigma + Easout	3.88	4.00	4.00
Stigma + water	4.00	3.88	4.00

LSD., (0.05) = 0.65

LSD., (0.01) = 0.87

DF=60, MS=0.2134

Fruit shape: 0=malformed fruit; 4=perfect shape.

Each figure represents the mean of four observations.

Captan 50% WP 2g l^{-1} and Easout 70% WP 1g l^{-1} .

(Fig. 20, 21 and 22).

There were significant differences between the cultivars. Bounty showed the least fruit malformation while Sparkle and Redcoat produced significantly more malformed fruit after the application of pollen treated with fungicides (table 10), (Fig. 23).

4.2.3. Number of Achenes

The total number of achenes was not significantly affected by fungicide application. However, application of pollen which had been sprayed with fungicides increased the number of aborted achenes. Captan had the most pronounced effect on achene abortion (tables 11, 12, 13).

Analysis of variance of transformed data (square root transformation), gave the the same results (table 14).

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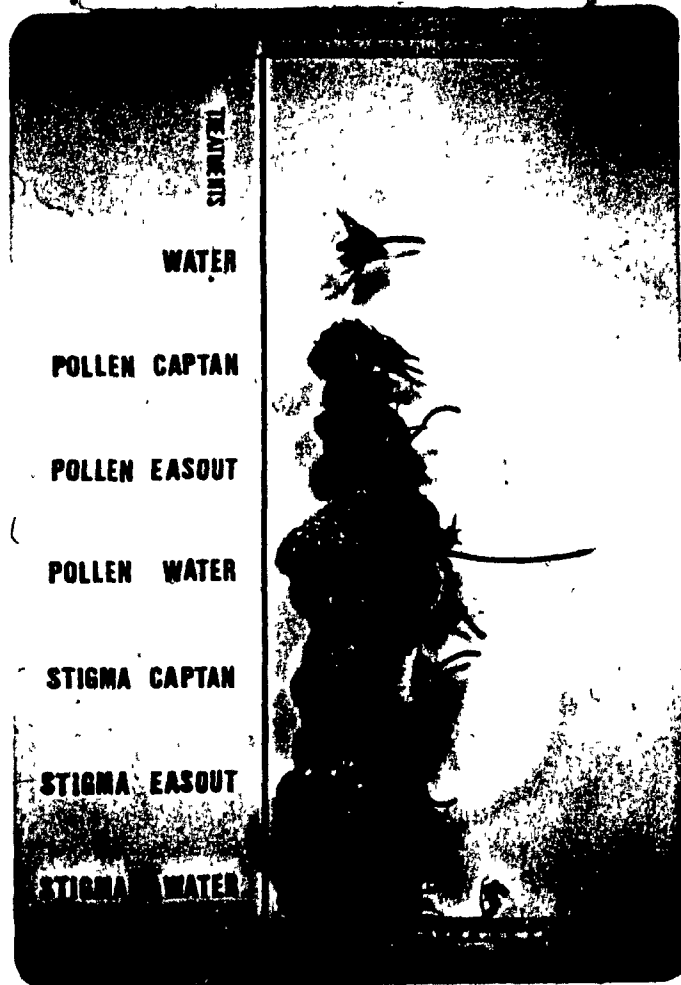


Figure 20. Fruits of the cv. Bounty following treatment of either pollen or stigma with fungicides.

Table 11. Berry shape and achene number (cultivar Bounty) following treatment of either pollen or stigma with fungicides. (All flowers were emasculated before treatment application).

	shape	achenes			
		total	aborted	sound	%sound
control-unpollinated	0.00	163	163	00	00
pollen + Captan	3.25	205	139	66	30
pollen + Easout	3.50	193	61	132	68
pollen + water	3.88	240	51	189	79
stigma + Captan	4.00	260	43	217	83
stigma + Easout	3.88	185	26	159	85
stigma + water	4.00	200	50	150	76
LSD. (0.05)	0.65	42	37	43	15
LSD. (0.01)	0.87	55	49	58	20
DF	60	60	60	60	60
MS	0.21	862	674	944	116

fruit shape: 0=malformed fruit, 4=perfect shape.

Captan 50% WP, 2g l^{-1} & Easout 70% WP, 1g l^{-1} .

Each figure represents the mean of four observations.

Fruit shape: 0=malformed fruit; 4=perfect shape.

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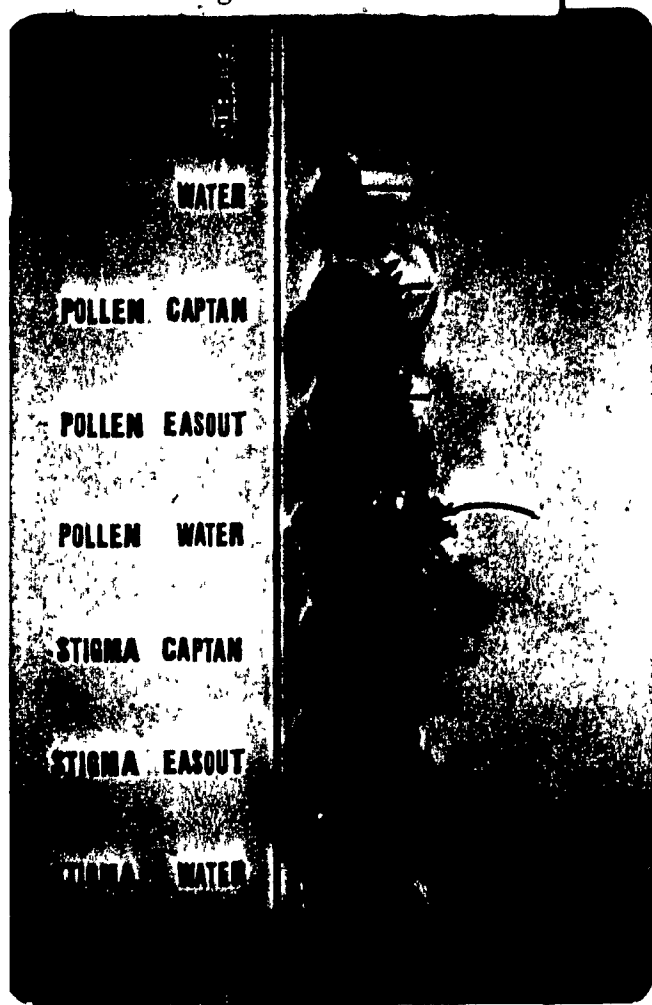


Figure 21. Fruits of the cv. Redcoat following treatment of either pollen or stigma with fungicides.

Table 12. Berry shape and achene number (cultivar Redcoat) following treatment of either pollen or stigma with fungicides. (All flowers were emasculated before treatment application).

	shape	achenes			
		total	aborted	sound	%sound
control-unpollinated	0.00	159	159	00	00
pollen + Captan	1.00	196	131	65	34
pollen + Easout	1.75	239	129	110	45
pollen + water	3.88	236	42	194	81
Stigma + Captan	3.88	198	39	159	79
stigma + Easout	4.00	208	39	169	81
stigma + water	3.88	183	34	149	82
LSD. (0.05)	0.65	42	37	43	15
LSD. (0.01)	0.87	55	49	58	20
DF	60	60	60	60	60
MS	0.21	862	674	944	116

Captan 50% WP, 2g l^{-1} & Easout 70% WP, 1g l^{-1} .
Each figure represents the mean of four observations.

Fruit shape: 0=malformed fruit; 4=perfect shape.

COLOURED PICTURES
Images en couleur

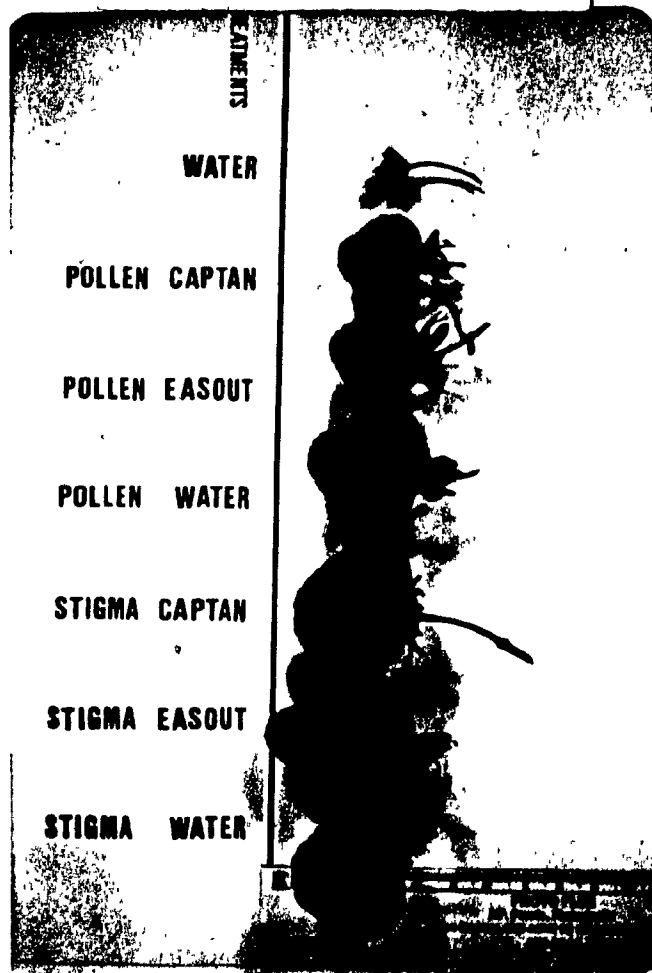


Figure 22. Fruits of the cv. Sparkle following treatment of either pollen or stigma with fungicides.

Table 13. Berry shape and achene number (cultivar Sparkle) following treatment of either pollen or stigma with fungicides. (All flowers were emasculated before treatment application).

	shape	achenes			
		total	aborted	sound	%sound
control-unpollinated	0.00	155	154	1	00
pollen + Captan	1.63	237	171	66	28
pollen + Easout	3.38	206	39	167	81
pollen + water	4.00	216	29	187	86
stigma + Captan	4.00	232	25	207	89
stigma + Easout	4.00	221	16	205	92
stigma + water	4.00	235	13	222	94
LSD. (0.05)	0.65	42	37	43	15
LSD. (0.01)	0.87	55	49	58	20
DF	60	60	60	60	60
MS	0.21	862	674	944	116

Captan 50% WP, 2g l^{-1} & Easout 70% WP, 1g l^{-1} .
Each figure represents the mean of four observations.

Fruit shape: 0=malformed fruit; 4=perfect shape.

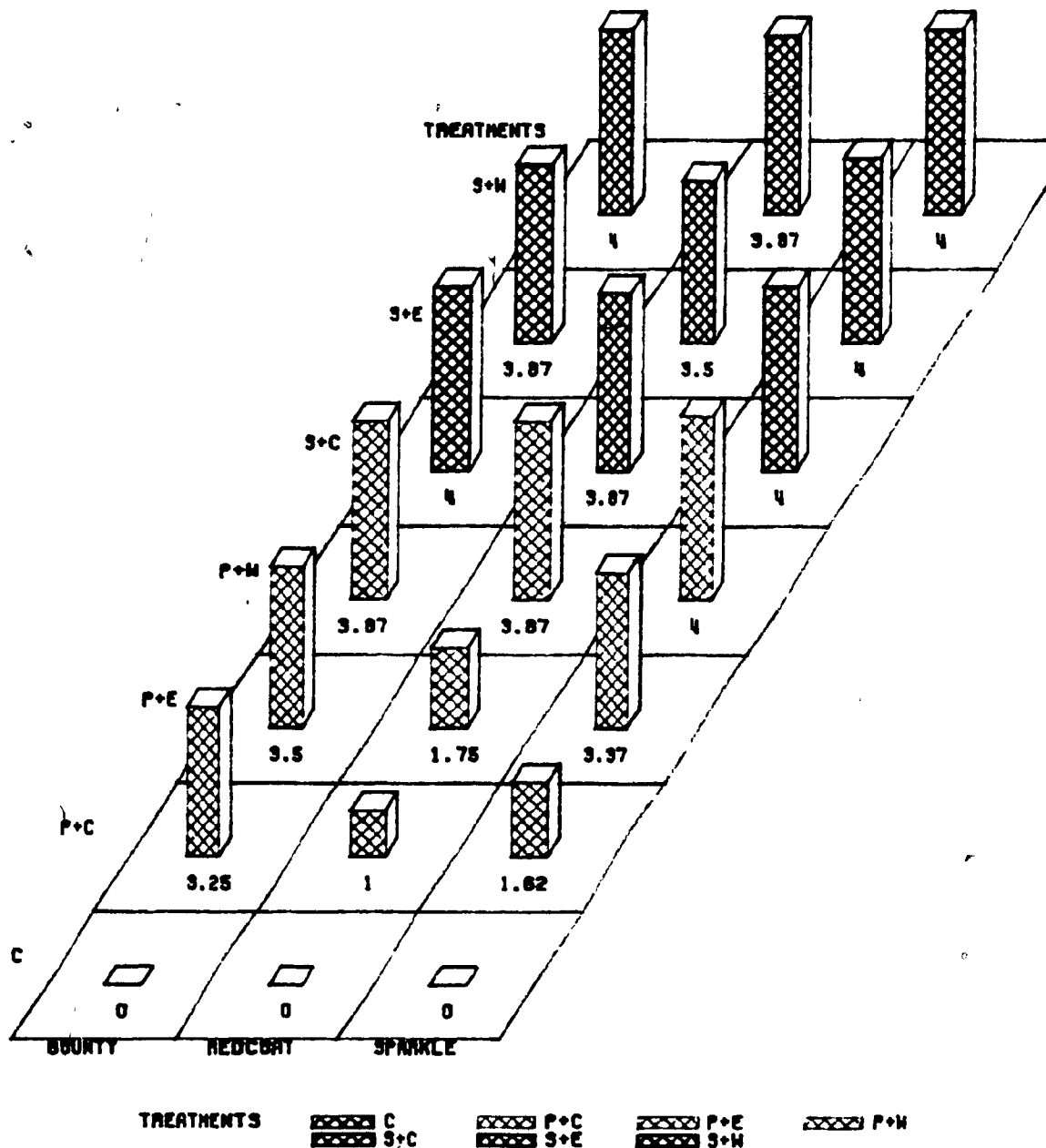


Figure 23. Shape rating of the fruit of three strawberry cvs. following treatment of either stigma or pollen with fungicides (0 = malformed fruit; 4 = perfect shape). Each bar represents the mean of eight observations. C = control P + C = pollen + Captan P + E = pollen + Easout S + C = stigma + Captan S + E = stigma + Easout S + W = stigma + water.

Table 14. Achene number of three strawberry cultivars (square root transformation) following treatment of either pollen or stigma with fungicides.
All flowers were emasculated before treatment applications

Treatments	cultivars					
	Bounty		Redcoat		Sparkle	
	aborted sound achenes		aborted sound achenes		aborted sound achenes	
Control-unpollinated	9.98	0.47	9.99	0.21	9.96	0.60
Pollen + Captan	8.25	5.34	8.08	5.80	8.47	5.29
Pollen + Easout	5.30	8.14	7.32	6.74	4.08	8.99
Pollen + water	4.33	8.89	4.13	8.98	3.36	9.31
Stigma + Captan	4.02	9.14	4.43	8.92	3.23	9.44
Stigma + Easout	3.70	9.26	4.27	9.01	2.66	9.01
Stigma + water	4.71	8.68	4.17	9.07	2.38	9.71
LSD., (0.05)	1.53	1.04	1.53	1.04	1.53	1.04
LSD., (0.01)	2.03	1.38	2.03	1.38	2.03	1.38
DF	60	60	60	60	60	60
MS	1.17	116.32	1.17	116.32	1.17	116.32

Captan 50% WP, 2gl^{-1} & Easout 70% WP, 1gl^{-1} .

Each figure represents the mean of four observations.

4.3. Experiment 3. Effects of Fungicide Concentration Incorporated into the Growing Medium on Pollen Germination and Length of Pollen Tubes

There was no significant interaction between the variables (fungicide and cultivar). When the effect of time was taken into account, a dependent effect of fungicide, cultivar and time was found on percent pollen germination at the 0.05 level of significance. This indicated that these factors were not independent in their effects on percent germination (see appendix. 1, table. 15) However, an independent effect of factors was found with respect to the length of pollen tubes (see appendix. 1, table.16).

There were significant differences between cultivars in terms of percent pollen germination in vitro after the first three hours of incubation (table 15). and after twenty four hours. The germination of Bounty and Redcoat pollen was significantly greater than Sparkle at the end of the test (table 15). A similar situation existed for the length of pollen tubes. The degree of inhibition of germination and pollen tube growth is shown in table 16. Figures 24 to 28 show the effect of fungicide concentration on pollen germination and pollen tube growth.

Captan at all concentrations and Easout at 2000ppm were very inhibitory while Easout at 500 and 1000ppm significantly less inhibitory (tables 15 and 16), (Fig. 25 & 26).

Table 15. Percent pollen germination of three strawberry cultivars treated with two fungicides over a range of concentrations in vitro, after 3 & 24 hours incubation.

Treatments	Incubation time					
	3 hours			24 hours		
	Bounty	Redcoat	Sparkle	Bounty	Redcoat	Sparkle
Control	5.22	4.37	5.70	7.52	6.97	5.82
E 500ppm	1.80	2.31	2.05	1.55	3.45	1.56
E 1000ppm	1.53	1.86	2.22	2.60	3.00	1.42
E 2000ppm	1.15	1.14	1.21	1.82	1.05	1.23
C 500ppm	1.84	1.00	1.34	1.55	1.62	1.68
C 1000ppm	1.31	1.00	1.24	1.00	1.05	1.18
C 2000ppm	1.27	1.00	1.10	1.14	1.24	1.00

LSD. (0.05) = 0.78 DF=126 MS=0.22

LSD. (0.01) = 0.10 DF=126 MS=0.22

All figures are square root conversions for statistical purposes

Each figure represents the mean of three observations.

C=Captan 50% WP a.i. & E=Easout 70% WP a.i.

Table 16. Length (mm) of the pollen tubes of three strawberry cultivars treated over a range of fungicide concentration in vitro, after 3 & 24 hours incubation.

Treatments	Incubation time					
	3 hours			24 hours		
	Bounty	Redcoat	Sparkle	Bounty	Redcoat	Sparkle
Control	0.71	0.76	0.76	1.03	1.01	0.93
E 500ppm	0.72	0.72	0.71	0.78	0.76	0.73
E 1000ppm	0.71	0.71	0.71	0.74	0.75	0.71
E 2000ppm	0.71	0.71	0.71	0.72	0.71	0.71
C 500ppm	0.71	0.71	0.71	0.72	0.72	0.72
C 1000ppm	0.71	0.71	0.71	0.71	0.71	0.72
C 2000ppm	0.71	0.71	0.71	0.71	0.71	0.71

LSD. (0.05) = 0.04 DF=126 MS=0.0005

LSD. (0.01) = 0.05

E=Easout 70% WP a.i. & C=Captan 50% WP a.i.

Each figure represents the mean of three observations.

Fig. 24 Pollen grains treated with 10%
sucrose solution after 24 hours
incubation.

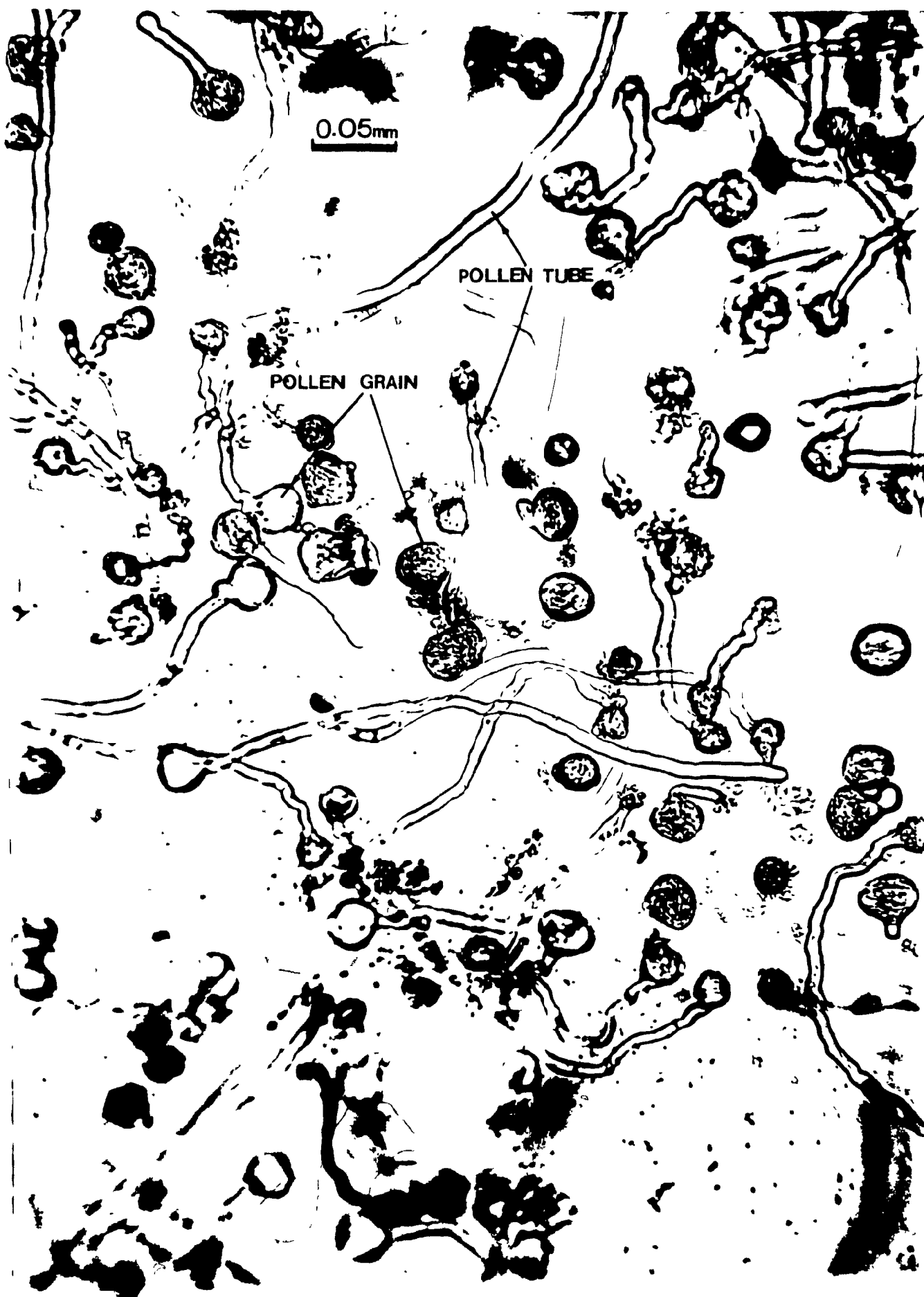


Fig. 25 Pollen grains treated with 500ppm
of Captan incorporated into 10%
sucrose solution after 24 hours
incubation.

0.05 mm

POLLEN GRAIN

POLLEN TUBE

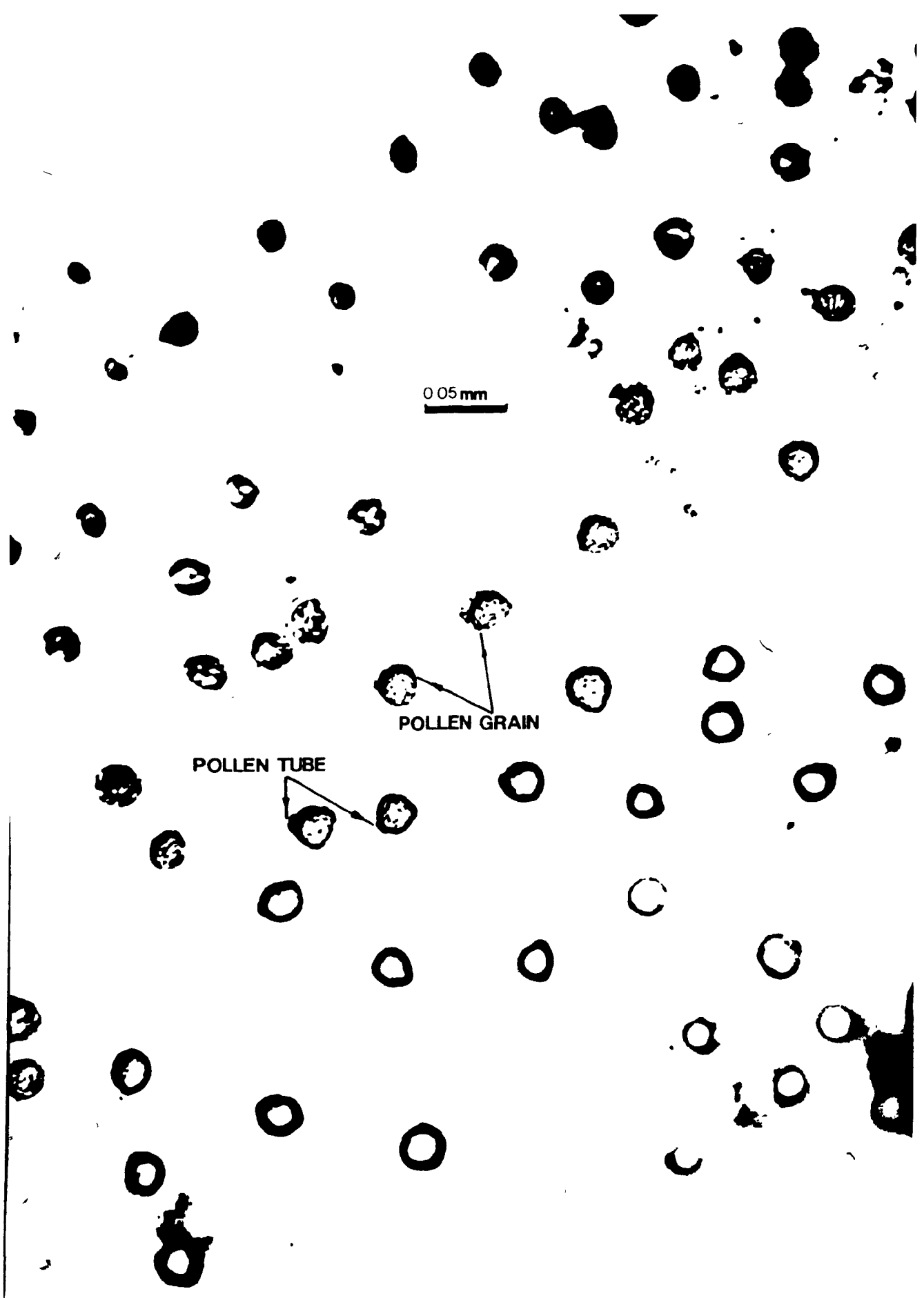


Fig. 26 Pollen grains treated with 500ppm of
Easout incorporated into 10% sucrose
solution after 24 hours incubation.

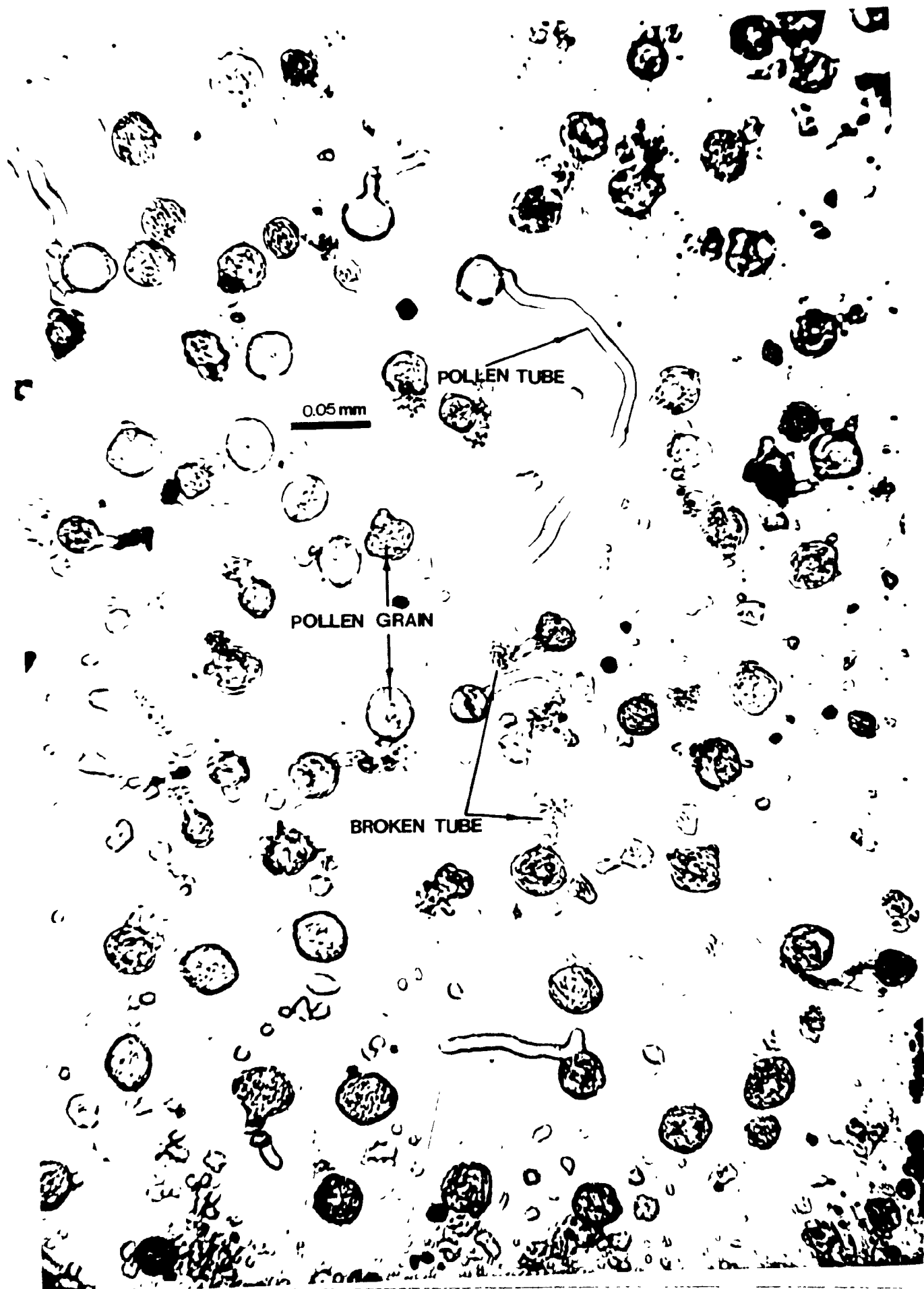


Fig. 27 Pollen grains treated with 1000ppm
of Easout incorporated into 10%
sucrose solution after 24 hours
incubation.

0.05mm

BROKEN TUBE

POLLEN TUBE

POLLEN GRAIN

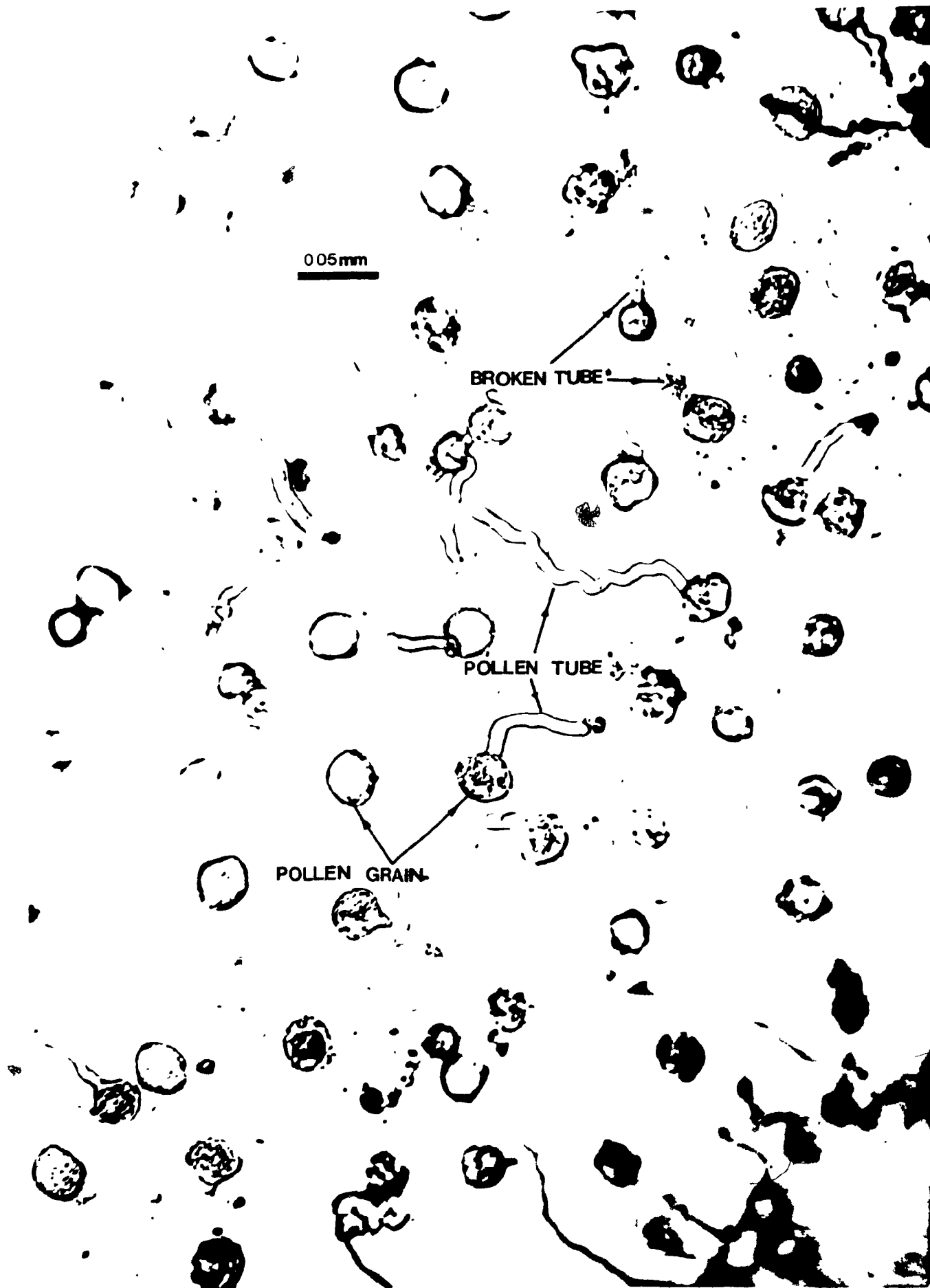
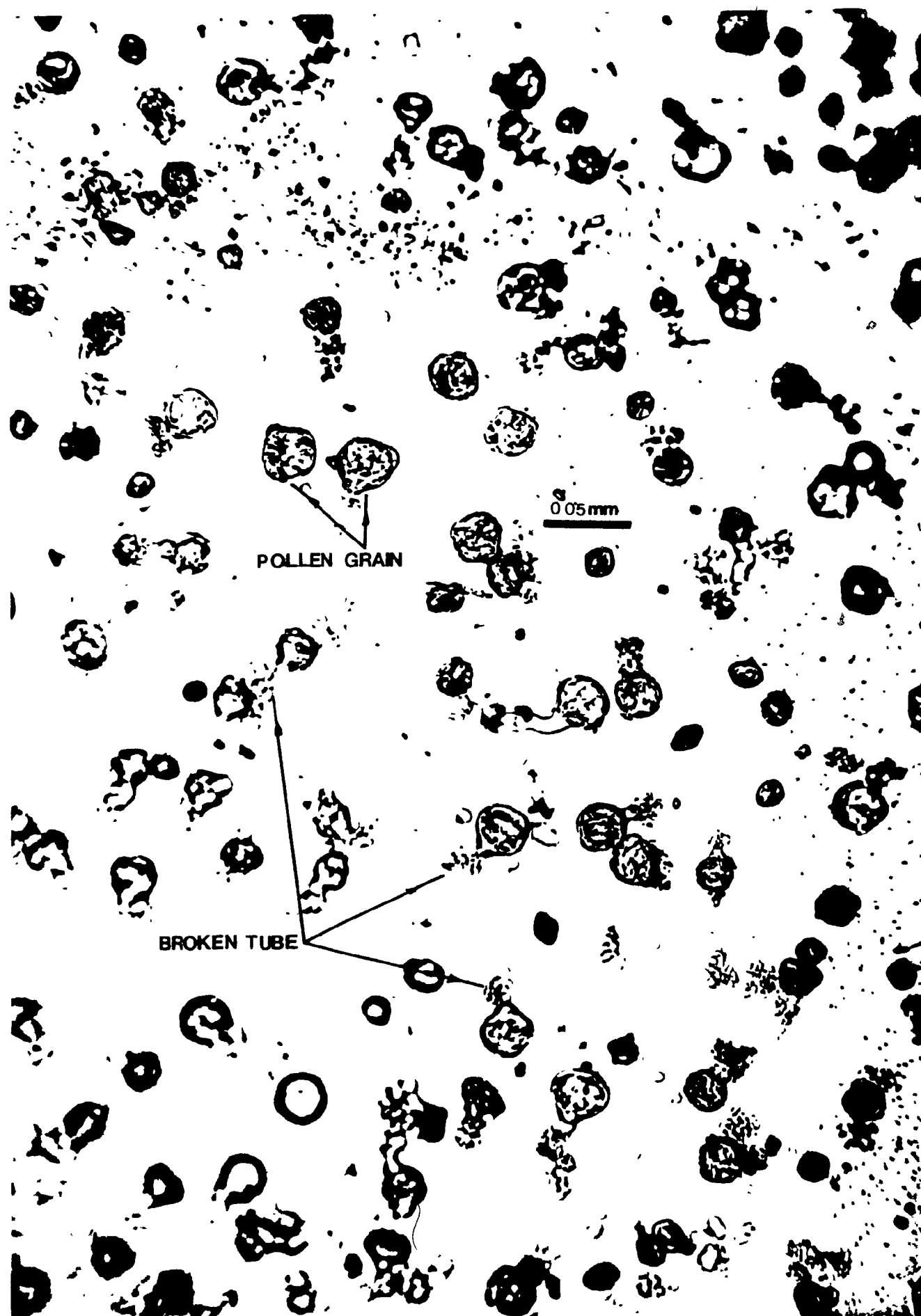


Fig. 28 Pollen grains treated with 2000ppm
of Easout incorporated into 10% sucrose
solution after 24 hours incubation.



POLLEN GRAIN

0.05mm

BROKEN TUBE

This difference was apparent after three hours incubation and persisted until measurements were stopped (twenty-four hours after incubation), (see appendix 4, Fig. 3 to 10). Easout at 500ppm was the least inhibitory (tables 15 & 16), (see appendix 4, Fig. 3 to 10).

According to the analysis of variance none of the fungicide treatments (Captan 500, 1000, 2000ppm and Easout 500, 1000, 2000ppm) were significantly different from each other with respect to their effect on pollen tube length during the first nine hours (see appendix 4, Fig. 4, 6, 8), but significant differences were apparent after twenty-four hours (see appendix. 4, Fig. 10). Captan at 500, 1000, or 2000ppm and Easout at 2000ppm had the most detrimental effect on the length of the pollen tube. There was no difference between Easout at 1000 or 2000ppm, or Captan at 500ppm, as was found with percent pollen germination. Easout at 500ppm was the least inhibitory to pollen tube growth of the fungicides tested (table 16).

When time of incubation was considered as a factor, there were no significant differences between cultivars in terms of pollen germination and pollen tube length (see appendix. 1, tables 15 & 16).

There was no difference between the effects of Easout at 1000 or 500ppm, on pollen germination or length of pollen tube after twenty-four hours incubation (Tables 15, 16). Similar results were obtained for percent pollen germination and tube length when data for all three cultivars was analysed together (table 17).

Table 17. Average percent pollen germination and pollen tube length of three strawberry cultivars treated with two fungicides over a range of concentrations in vitro, after 3 & 24 hours incubation.

Treatment	Time			
	3 hours		24 hours	
	% pollen germination	tube length	% pollen germination	tube length
Control	5.10a	0.76a	6.77a	0.99a
E 500	2.05b	0.72b	2.56b	0.76b
E 1000	1.87c	0.72b	2.34bc	0.73c
E 2000	1.16d	0.71b	1.37d	0.71cd
C 500	1.39cd	0.71b	1.61cd	0.71cd
C 1000	1.18d	0.71b	1.08d	0.71d
C 2000	1.12d	0.71b	1.13d	0.71d

Duncan's new multiple range test.
means with the same letters are not significantly different from each other at 0.05 level of significance.

All figures are square root conversions for statistical purposes.

E=Easout 70% WP, & C=Captan 50% WP
each figure represents the mean of three observations.

4.4. Experiment 4. Effects of Fungicides on Berry Quality and Yield and the Role of Insect Pollinators

There was no significant interaction between cultivars and fungicides. See appendix 1, tables 17 to 23.

4.4.1. Fruit Number and Fruit Weight

The application of fungicides did not influence fruit number or weight. Comparing the cultivars, Bounty gave the highest yields (table 18), (Fig. 29).

Table 18. Fruit number per plant (plants with multiple crowns) and mean individual fruit weight (g) of three strawberry cultivars treated with two fungicides.

factors	cultivar			fungicide		
	Redcoat	Sparkle	Bounty	Captan	Easout	water
Fruit No	189b	227b	364a	259a	268a	254a
Fruit weight	1.448b	1.003c	2.303a	1.54a	1.66a	1.55a

Duncan's new multiple range test.

Means with the same letter are not significantly different from each other at 0.05 level of significance. (For three levels of the same factor).

Captan 50% WP, 2g l⁻¹ & Easout 70% WP, 1g l⁻¹.

Each figure is the mean of 12 observations.

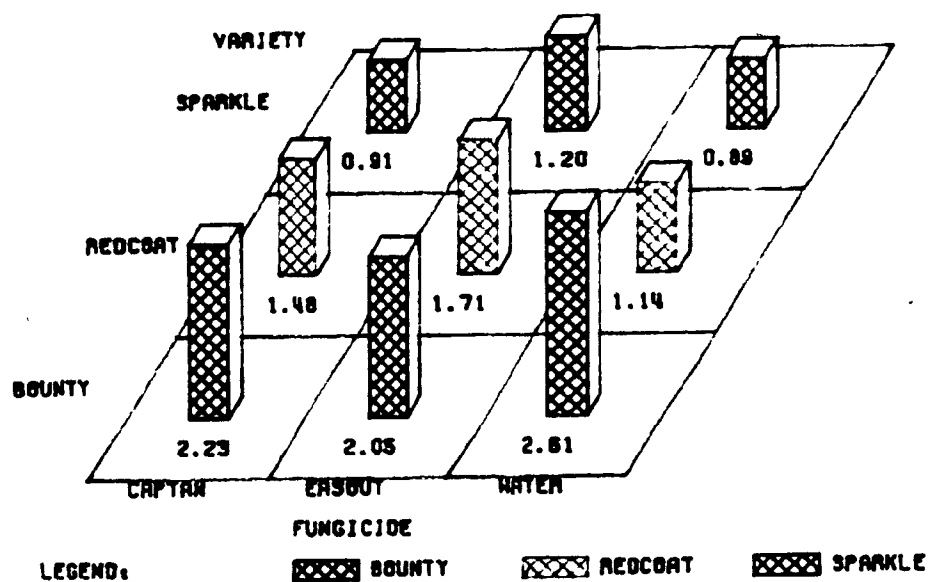


Figure 29. Mean and individual fruit weight (g) of three strawberry cvs. treated with fungicides during bloom. Each bar is the average fruit weight of four observations.

4.4.2. Fruit Shape and Disease

There was no difference between cultivars in terms of fruit disease (table 19). Bounty produced significantly better shaped fruits and Sparkle the worst shaped fruits (table 19). Easout and Captan significantly increased the number of malformed berries (Fig. 30) but reduced the number of diseased fruits (Fig. 31), (table 19).

Table 19. Fruit shape and disease rating of three strawberry cultivars treated with two fungicides.

factors	cultivar			fungicide		
	Redcoat	Sparkle	Bounty	Captan	Easout	water
Fruit shape	2.49b	2.11c	3.36a	2.49b	2.47ab	2.80a
Fruit disease	1.00	0.92	0.50	0.17b	0.50b	1.75a

Duncan's new multiple range test.

Means with the same letter are not significantly different from each other at 0.05 level of significance, (For three levels of the same factor).

Captan 50% WP, 2g l⁻¹ & Easout 70% WP, 1g l⁻¹.

Each figure is the mean of 12 observations.

Fruit shape: 0=malformed fruit, 4=perfect shape.

fruit disease: 0=no disease, 4=totally infected.

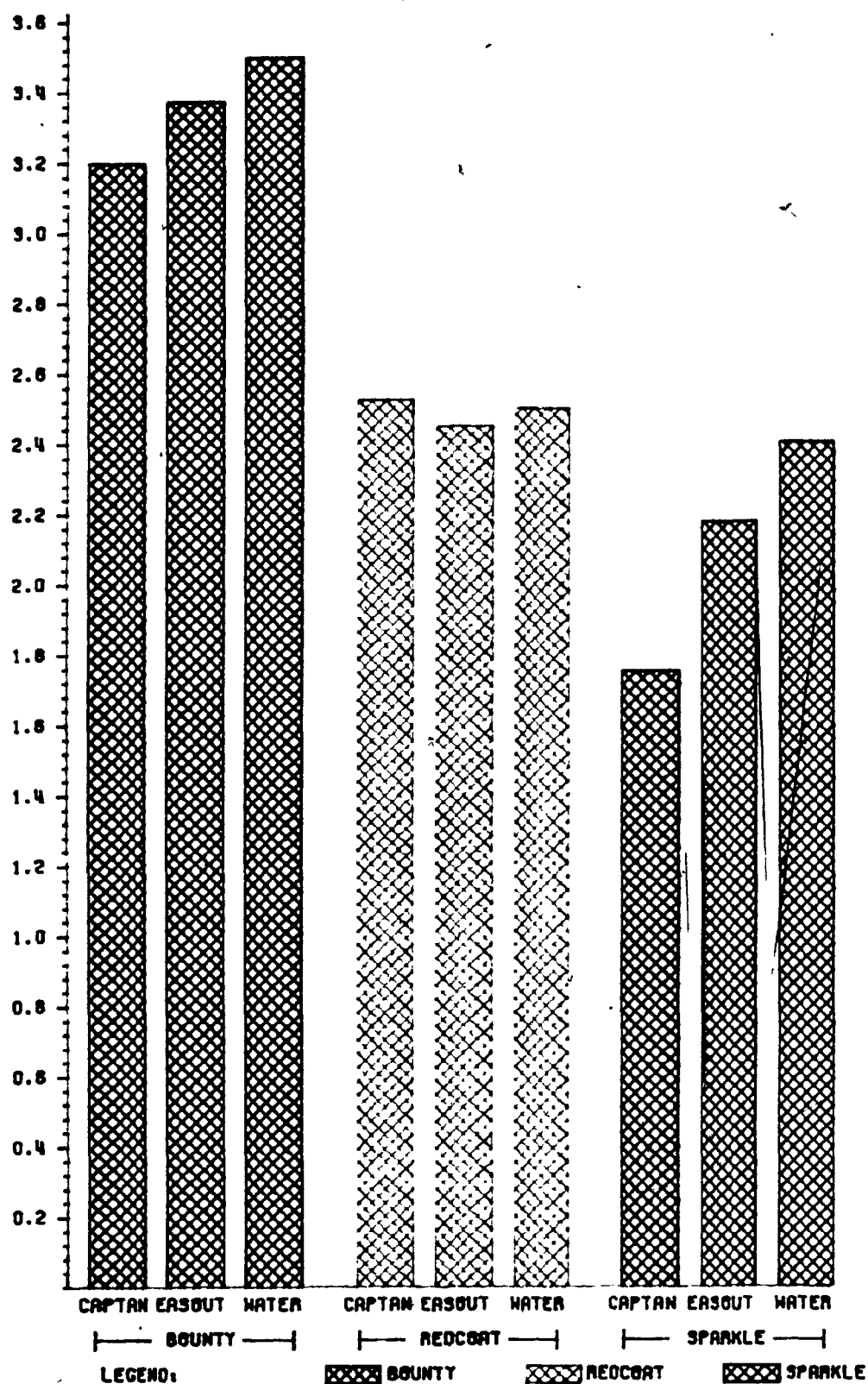


Figure 30. Fruit shape rating of three strawberry cvs. treated with fungicides during bloom (0 = malformed fruit; 4 = perfect shape). Each bar is the average of four observations.

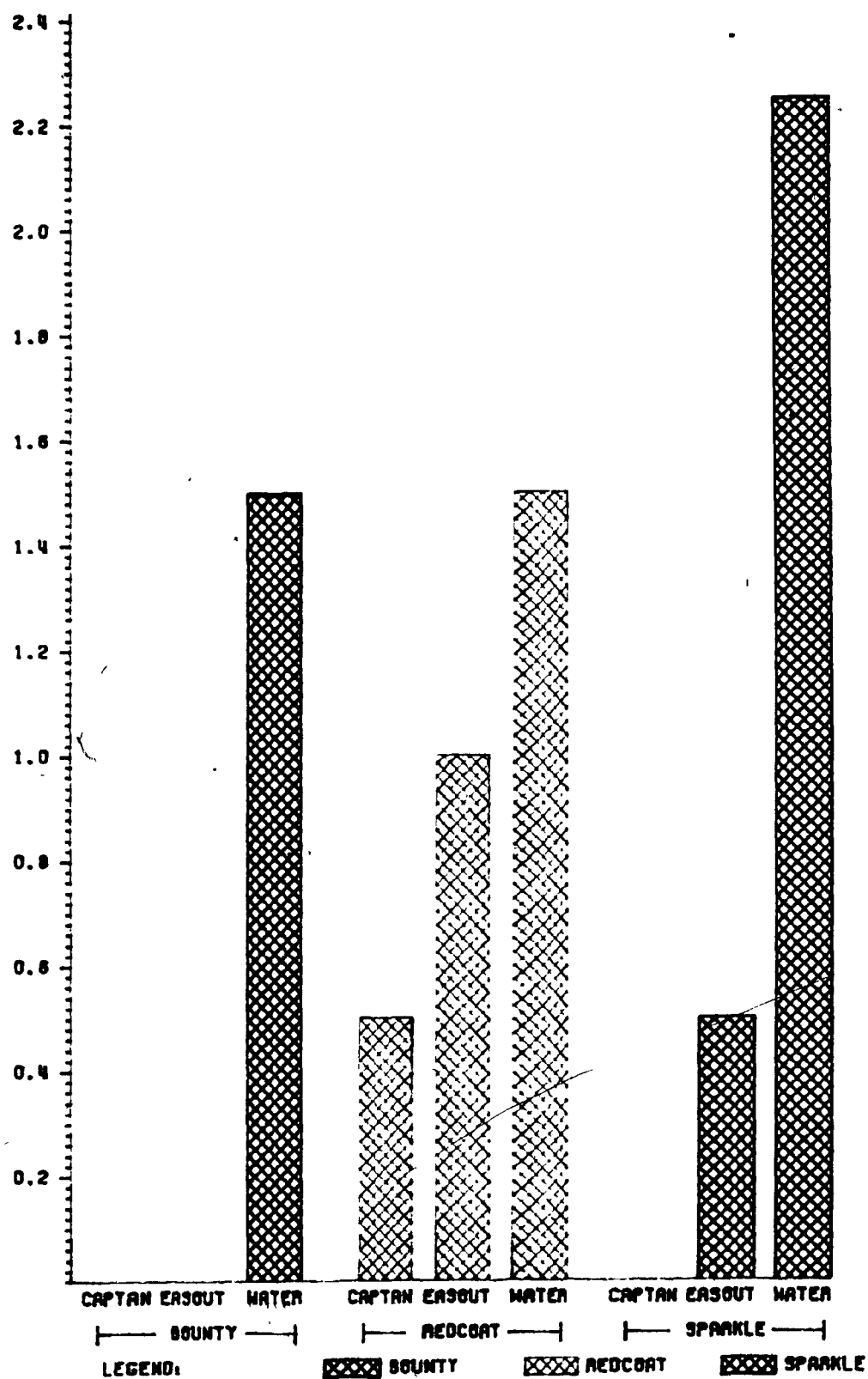


Figure 31. Fruit disease rating of three strawberry cvs. treated with fungicides during bloom period (0 = no disease; 4 = totally infected). Each bar is the average of four observations.

4.4.3. Number of Achenes

There were no differences between cultivars in terms of total, aborted and sound achenes, even when the data were transformed to percentages or square roots (table 20).

Fungicide application had no effect on total and sound achenes (see table 20) but Captan significantly increased the number of aborted achenes. However, after the data were transformed to percentages or square roots, the effects of the fungicides, especially Captan, on the reduction of the number of sound achenes were apparent (table 20), (Fig. 32).

Table 20. Achene number of three strawberry cultivars treated with two commercial fungicides.

factors	cultivar			fungicide		
	Redcoat	Sparkle	Bounty	Captan	Easout	water
aborted achenes	69	114	89	110a	68b	96b
sound achenes	227	257	292	236 ^a	261	278
total achenes	326	341	381	346	329	374
percent sound achenes	80	67	75	68	79	74
percent sound ** achenes	8.9	8.1	8.7	8.2b	8.9a	8.6a

** Square root conversion for analysis of variance procedure.

Duncan's new multiple range test, (For three levels of the same factor. Means with the same letter are not significantly different from each other at 0.05 level of significance.
 Captan 50% WP, 2gl⁻¹ & Easout 70% WP, 1gl⁻¹.
 Each figure is the mean of 12 observations.

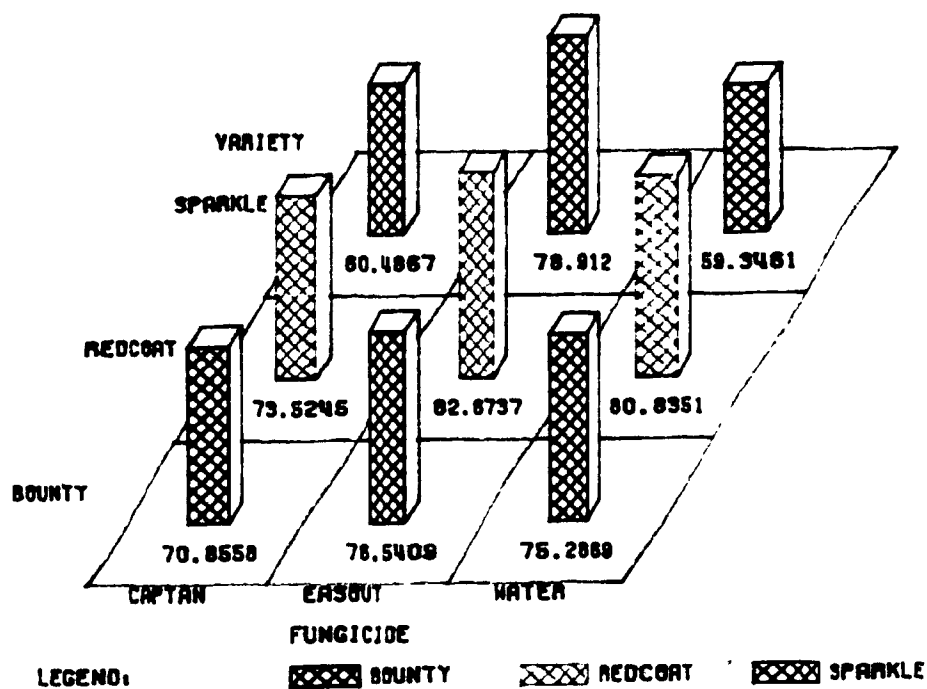


Figure 32. Percentage of sound achenes of three strawberry cvs. treated with fungicides during bloom period. Each bar is the mean of four observations.

4.4.4. Observations on the Insects Visiting Strawberry Flowers

The results of the field experiment in summer 1981 indicated that the deleterious effects of fungicide application during the bloom period might be overcome by insect pollinators, especially honey bees (Fig. 33).

To investigate the activity of insects in the field, the same field was used. Eight rows of strawberry plants were randomly selected and a 1.5 m length of each row was used for counting the number of insects that visited each flower during 15 minute periods. The results, which are the mean of eight rows, over a period of ten days, during the bloom period, are summarized in table 21.

Some insects were collected for identification and are shown in Fig. 34, 35, 36, 37, 38, 39.

Table 21. Observations of the insects visiting strawberry flowers.

type	Average time on flower (Seconds)	Number observed in 15 minute period (Mean of 80 observations)
Honey bee	15	40
Bumble bee	5	1
Solitary bee	10	10
Fly	10	4
Butterfly	8	1
Other	7	3

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Figure 33. Pollination of strawberry flowers by a honey bee, Apis mellifera, summer 1981.

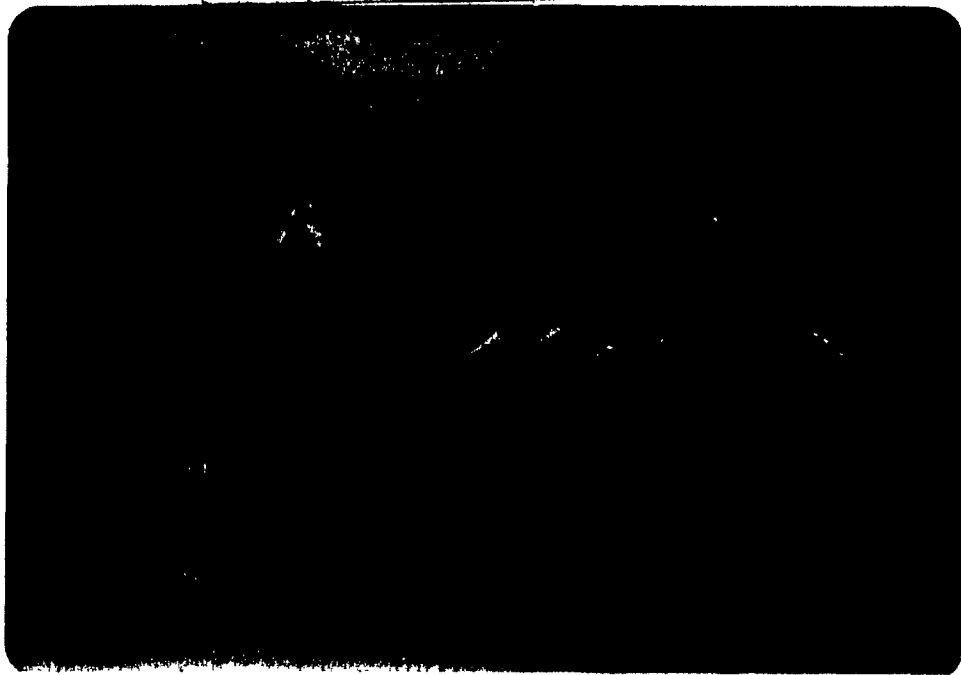


Figure 34. Bumble bees, unidentified.

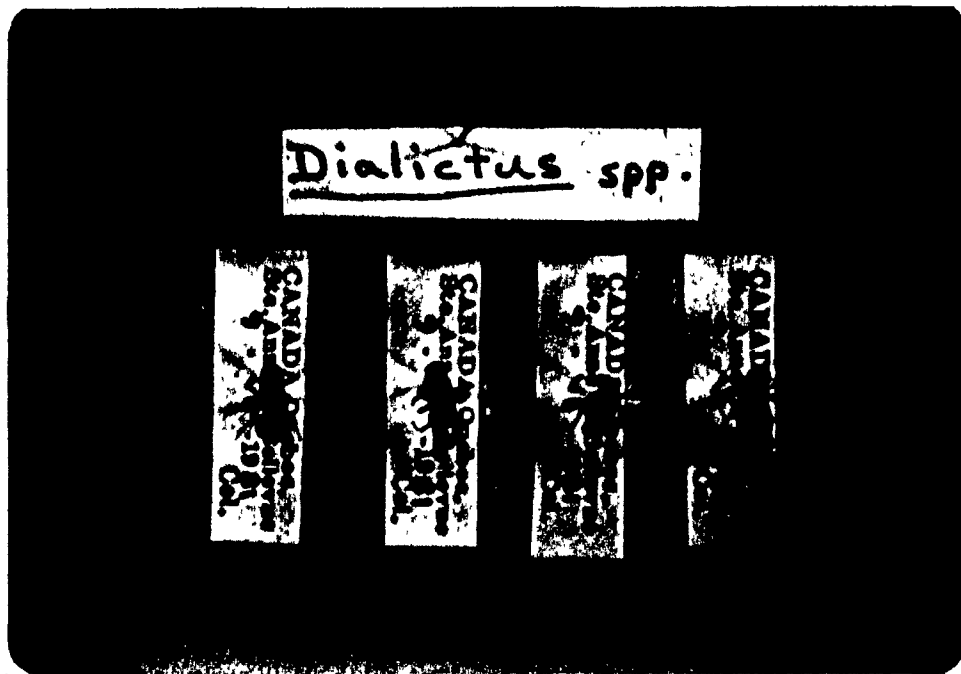


Figure 35. Solitary bees, Dialictus spp.

Insects collected from strawberry flowers during the blossom period, 1982.

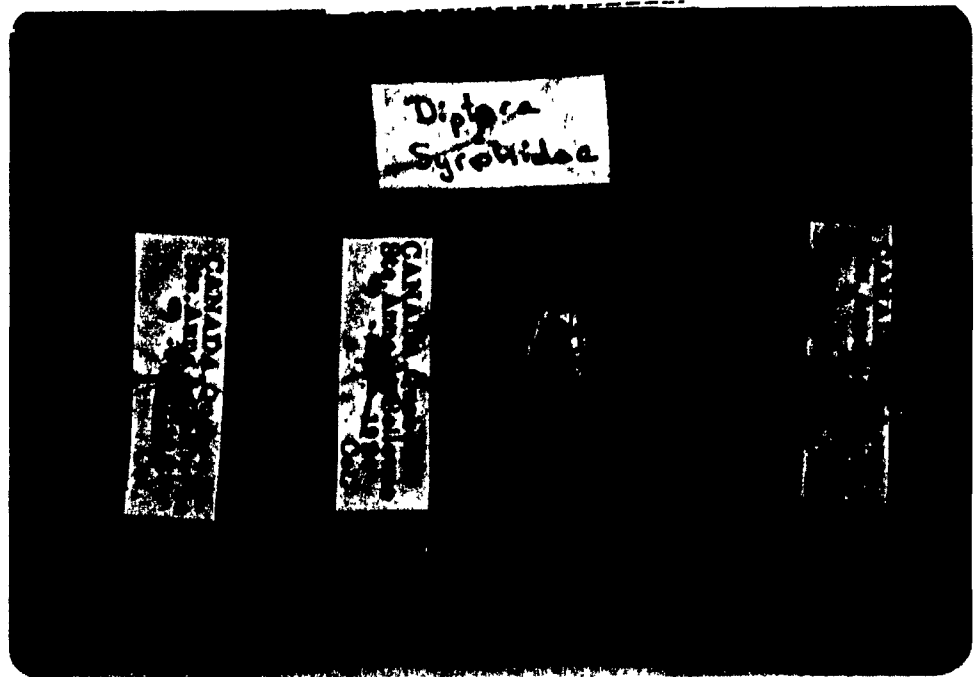


Figure 36. Flies (Diptera), unidentified.



Figure 37. Lady bugs, unidentified.

Insects collected from strawberry flowers during the blossom period, 1982.

Andrenidae
Andrena spp.



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Figure 38. Solitary bees (Andrenidae).

Crabronidae
Hestica
confluente
(Say)



Figure 39. Solitary bee (Crabronidae).

Insects collected from strawberry flowers during the
blossom period, 1982.

5. DISCUSSION

5.1. The Effects of the Two Fungicides Used on Strawberries

The aim of this study was to determine whether commercial fungicides applied during flowering could affect strawberry fruit weight and quality.

Captan and Easout were used, and it is possible that other ingredients in their formulations may themselves affect pollen germination (Gentile et al., 1973, Shivanna, 1972). In addition, the method of fungicide application employed in this study (i.e. spraying until "run-off") probably deposited more fungicide on the anthers than is normally the case in commercial production and hand pollination may have deposited so much pollen on the stigma that, even if germination was partly inhibited by the fungicide, there may still have been sufficient viable pollen to allow fertilization.

5.1.1. Beneficial Effects

Fungicides and time of application had no effect on flower and fruit number, since flowers were already initiated in the previous year. Fruit rots were satisfactorily controlled by application of either fungicide, results were similar to Bennett's findings (1972) that fungicide applications reduced fruit rot (B. cinerea). but neither of the fungicides reduced berry size or caused visible phytotoxicity in his trials.

The results confirm Eaton's findings (1963). His data indicated that each of the fungicides he used (applied during the bloom period) could reduce pollen germination and subsequently fruit weight but he did not consider the reduction to be significant in the field. Furthermore he recommended the use of several fungicides for control of apple scab, (Venturia inaequalis Cke.), during bloom.

The results of the field experiments suggest that the relationships reported by Braun and Schonbeck (1965) between Captan and reduced apple fruit set, by Molnar (1972) between spraying fungicides and reduced apple yield and by Kaspars (1965) between spraying Captan and reduced cranberry fruit set and yield, do not seem to hold for the strawberry probably due to the self-fertility of strawberry flowers and their structure. However, the results do agree with the recent finding of Bristow (1981) that fruit set and berry development were not affected by fungicide application in the field and with the results of Howard and Albergt (1982) who reported that none of the fungicides in their field trial caused visible toxicity or affected fruit weight and all of them could control the incidence of fruit disease. In addition, the findings also agree with the results of Redalen (1980) who reported that, in the field, no significant differences in fruit set could be demonstrated for raspberries after application of Benomyl, Captan or Dichlofloanid, thus any of them could be applied for disease control.

3.1.2. Detrimental Effects

In this study fungicide applications always reduced the percentage of sound achenes. Lockhart (1967) reported that lowbush blueberries in plots treated with fungicides were not significantly different from those in control plots with respect to the seed count. This difference between strawberries and blueberries could be due to differences in the floral structure of the species. Strawberries have open flowers while the stamens of blueberry flowers are protected by their petals and pendant position.

These experiments confirm the results of Bennet (1968) who reported that application of fungicides to strawberry flowers increased the proportion of misshapen fruit. This may be because of reduced pollen germination as Eaton (1963) demonstrated with apple pollen.

In the field experiments fungicides had no adverse effect on percent fruit set or yield even though they appeared to be capable of increasing the number of aborted achenes and malformed berries.

The results presented here apparently contradict previous findings that aborted achenes and subsequent fruit weight are negatively correlated (Nitch, 1950, Eaton, 1966, Hall et.al, 1965). Why the percent fruit set and yield were not reduced despite the increase in the number of aborted achenes might be explained in two ways: one relates to

stimulation of the ovary and the other relates to compensation by the remaining sound achenes. For some fruits it has been demonstrated that an extract of pollen can stimulate fruit formation by unpollinated flowers (Nitch, 1952). Investigations have shown that this stimulation is mainly due to auxin (Nitch, 1952). This phenomenon may be accentuated under relatively higher temperatures. In this experiment it is possible that fungicide treated pollen (transferred by pollinators or wind to the stigma), while incapable of successful fertilization, might still have stimulated the release of some growth substances. Consequently, despite their being aborted achenes some increase in berry size may have resulted. In addition to this it must be realized that despite fungicide application there were some sound achenes on the berries. These sound achenes appear to be associated with a phenomenon of compensation. To fully understand this phenomenon it might be helpful to explain why there were any sound achenes despite fungicide application.

Untreated pollen (and therefore normally viable) may have been transferred to the treated plants from untreated plants by pollinators or wind or it may have come from the treated plants themselves. It has been shown that application of fungicides to flowers before anthers dehisce does not effect pollen germination (Eaton and Chen, 1969a; Church and Williams, 1977). Since strawberry flowers are highly self fertile, any pollen from such undehisced anthers could thus later successfully fertilize the ovules.

In any case, the presence of viable pollen and subsequent successful fertilization resulted in sound achenes capable of promoting normal fruit development. Small numbers of sound achenes can stimulate proportionately more development of the receptacle in terms of flesh weight per achene than larger numbers (Eaton and Chen, 1969b). Thus, the sound achenes on each berry could compensate for the aborted achenes and help to overcome the reduction in fruit size. It appears that a certain percentage of sound achenes are necessary for optimum yield. In appendix 4 (Fig. 11), it can be seen that as the number of sound achenes increases so does the fruit weight but the ratio "weight:number of sound achenes" decreases. This seems to indicate that while the number of sound achenes increases, each one contributes proportionately less to the weight. This said, it appears that this compensation is not extended to overcome malformation of the berries in most cases. The reason for this may be due to the uneven distribution of aborted achenes on the surface of the receptacle.

A combination of the stimulation effect and the phenomenon of compensation could account for the insignificant reduction in percent fruit set and yield.

5.2. Factors Affecting Response to Fungicide Application

Several factors may reduce the adverse effects of fungicide application as demonstrated in the field, greenhouse and laboratory experiments. These are:

5.2.1. Stigma Receptivity

Strawberry pistils may remain receptive for several days in the field in normal conditions (Moore, 1964). Damage to pollen (dehiscid anthers) at this time, due to fungicide application, may not severely affect the pollination and subsequent fruit weight because of the abundance of undehisced anthers. Flowers could remain receptive for even longer periods of time in cool weather (Eaton and Chen, 1969b). (The cool temperature tends to slow down the physiological aging of pistils (Moore, 1964) thus resulting in extension of pistil receptivity time.)

5.2.2. Cultivar

Response to the application of fungicides is cultivar dependent, Redcoat, Sparkle and Bounty showed significant differences in terms of fruit weight, number of achenes, fruit shape and disease. In all cases Bounty produced the greatest fruit weight, best fruit shape, minimum number of aborted achenes, and the least number of diseased fruit. Bounty's greater fruit weight, as compared with Sparkle and Redcoat,

might be because it also had the greatest number of flowers per plant. In some cases even two inflorescence per plant were found. Bounty is resistant to B. cinerea fruit rot (Craig, 1982) and has stiff peduncles which prevent the fruit from touching the soil which could account for Bounty having the least number of diseased fruit. The flowers of Bounty are more showy than the other two cultivars and this might have attracted more pollinators which resulted in optimum pollination by insects and subsequently better fruit shape. Bounty might be considered the most suitable cultivar for this region.

5.2.3. Time of Application

According to the results (table.3) early and late fungicide application did not significantly differ from each other in terms of their effect on percent fruit set, fruit weight, fruit disease and total of aborted achenes. However, early fungicide application increased the number of malformed berries. The malformation of those berries was probably because earlier in the season some of the flowers could have been damaged by the cold weather and fewer insects were available to pollinate flowers early in the blossom period. Consequently the pollen was unevenly distributed amongst the stigmas, with a subsequent uneven distribution of sound achenes, giving rise to malformations.

5.2.4. Concentration and the Type of Fungicide

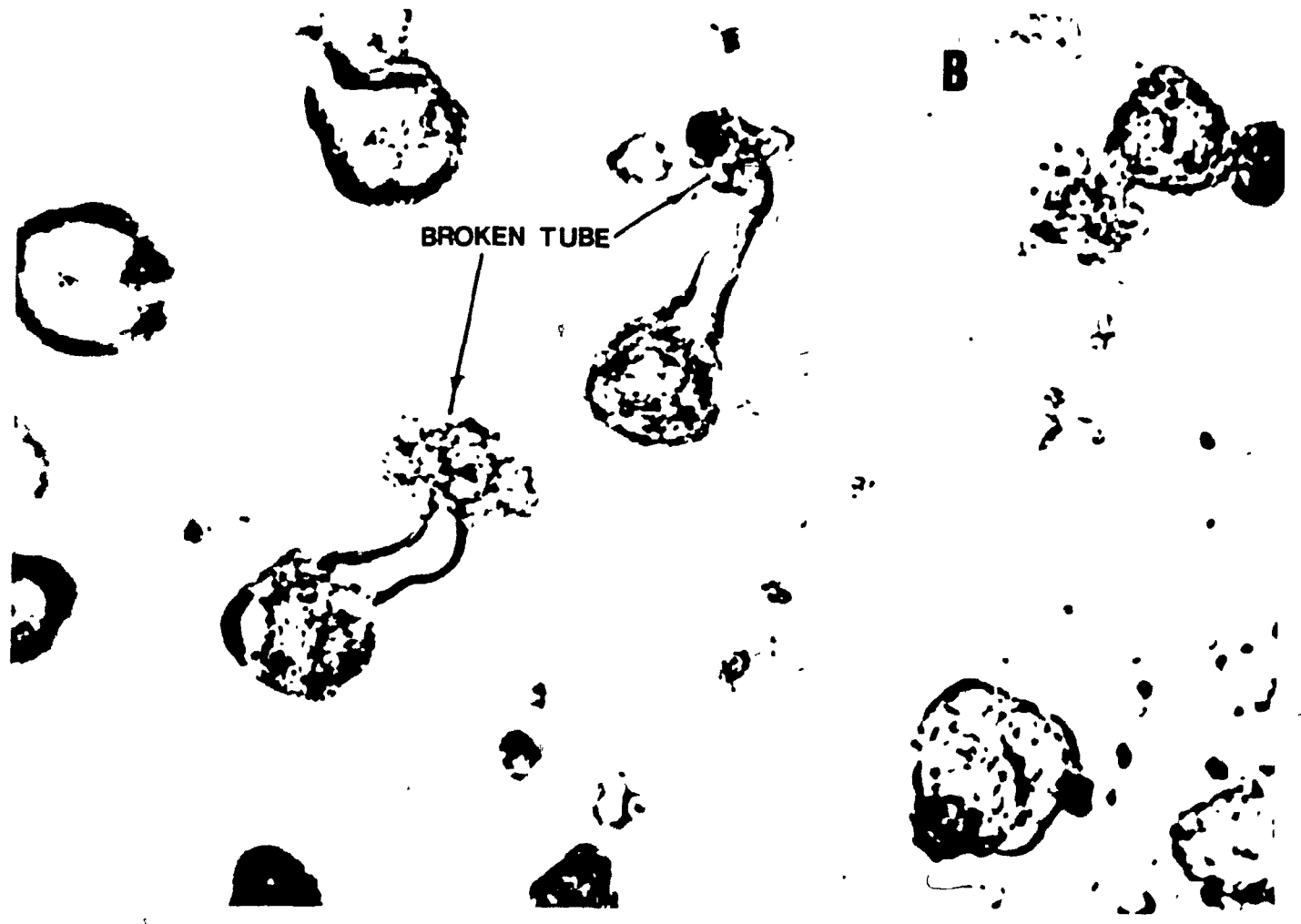
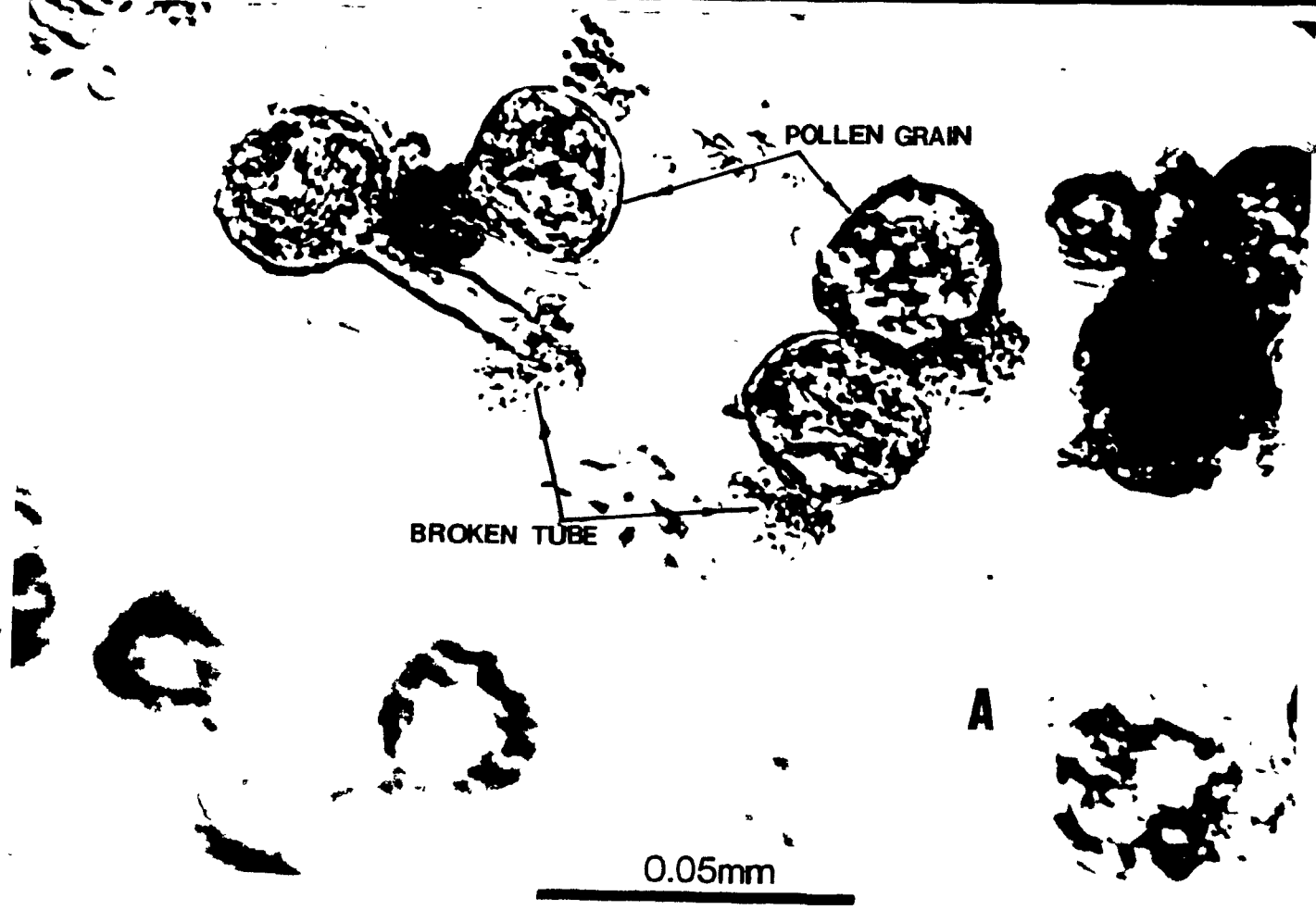
The results of experiment 2. showed that the fungicides (Captan and Easout) could inhibit pollen germination. Captan 50% WP and Easout 70% WP at rates of 500, 1000, 2000ppm were tested in vitro. Each fungicide was incorporated into a sucrose medium in which the pollen grains were allowed to germinate. Of the two fungicides tested, 500ppm of Easout 70% WP was least toxic and Captan 50% WP (500, 1000 and 2000ppm) was the most toxic to pollen. These results confirmed those of Rich (1957), Gartel (1961), Braun and Schonbeck (1963), Cristoferi et al. (1966), Kwack and Macdonald (1966), Eaton and Chen (1969b), Dancs and Kiss (1970) and Burth and Ramson (1974) who have demonstrated that many fungicides inhibit pollen germination in vitro when added to the medium. However, these findings may exaggerate the damage to be expected in terms of fruit set and yield in the field.

Captan almost entirely prevented pollen germination when incorporated into germination media at any concentration. Easout did not reduce pollen germination as severely as Captan but it did cause the pollen tubes to rupture after germination in most cases (Fig. 40).

The results suggest that the germination of strawberry pollen was arrested when in direct contact with fungicides. There were no significant differences between Captan (500, 1000, 2000ppm) and Easout (2000ppm) in this respect. However, Easout (500, 1000ppm) incorporated

Fig. 40 Pollen grains treated with 10%
sucrose solution and Easout.

- (A) 500ppm
- (B) 1000ppm



into media was less damaging than Captan (500, 1000, 2000ppm) or Easout at the higher rate.

It seems that the in vitro method of placing the pollen in fungicide solutions was too severe as the fungicides probably do not come into contact with pollen grains in this manner under natural conditions. Nonetheless the in vitro pollen germination test could be used to develop a standard test for fungicidal toxicity.

There were also differences in the eventual germination of pollen grains when slides were held for varying intervals of time (up to 24 hours). These differences indicate that, without being standardized this method is not suitable for investigating the adverse effects of fungicides on pollen germination. Also, the validity of these results in the field is questionable since, the ovules could remain receptive for up to 168 hours (Moore, 1964) in the field, the ovules could remain receptive for up to 168 hours (Moore, 1964).

5.2.5. Pollinating Insects

The abundance of pollinators was one of the most important factors in overcoming the adverse effects of fungicides in the field. Honey bees appeared to be the most frequent visitors of strawberry flowers and most of them collected pollen rather than nectar. Results suggest that in 1981 and 1982 the adverse affect of fungicides could have been mitigated by the pollinating insects.

There was no difference between the caged and uncaged plants in terms of fruit weight, percent fruit set and fruit shape. This could be due to the number of houseflies inside the cages which came out of the ground (Fig. 41). The presence of houseflies in the cages may have resulted from the dumping into the soil of left-over crops from the previous year. A significant infection of the berries by *B. cinerea* was found among the caged strawberries perhaps because of higher humidity or lower light intensity within the cages.

5.3. Susceptibility of Floral Parts to Fungicide Damage

Spraying anthers with Captan after dehiscence significantly reduced berry weight and number of sound achenes. This confirms the results of Eaton and Chen (1969b) who demonstrated that Captan acts directly on pollen germination, not on the receptivity of the stigma. These results confirm the findings of Braun and Schonbeck (1965), they concluded that none of the female parts of apple flowers were damaged by Captan. Further Schmidt (1956) showed that while Captan did not damage the stigma it did damage the pollen. The results presented here indicate that direct contact of strawberry pollen with Captan inhibited pollen tube development. When pollen tube development is inhibited there is no fertilization and a greater number of achenes abort, potentially reducing fruit weight.

When the pistils alone were sprayed with fungicide, there was no

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Fig.41. A strawberry flower (Cv. Bounty) being
visited by an unidentified Diptera sp.

effect on weight, number of sound achenes or percent sound achenes. Thus only the male parts of the strawberry flowers were damaged by fungicides. However, spraying the stigma with fungicide or tap water reduced berry weight more than when pollen was sprayed with tap water. It appears that applying any solution, no matter whether fungicide or water, reduces the receptivity of the stigma. It is possible that the fungicide or water washed-off some materials from the stigmatic surface which were essential for pollen germination. This needs further investigation since it could also reduce berry weight.

6. CONCLUSIONS

Despite a positive correlation between achene number and receptacle size, shown in all experiments (Appendix 4, Fig. 12), the relationship was not simple and it seems that only a certain percentage of achenes are necessary for full fruit development. Both fungicides reduced pollen germination in vitro and in vivo but in field conditions did not cause any significant yield reduction; whilst satisfactorily controlling the incidence of disease. The long receptive period of strawberry flowers and the deposition of viable pollen from untreated anthers, by foraging insects, could explain this apparent contradiction.

In conclusion, the use of fungicides during the flowering period offers good control of fruit rots and minimal risk of yield or quality reductions in the field.

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APPENDIX 1
ANALYSIS OF VARIANCE TABLES

Table.1

SUMMER 1981

General Linear Model Procedure

Dependent Variable: Fruit Number

<u>Sources</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>Mean-Square</u>
Model	143	439.8263	3.0757

<u>Sources</u>	<u>DF</u>	<u>Type IV SS</u>	<u>F Value</u>	<u>Pr > F</u>
cage	1	9.1003	1.14	0.3639
block	3	3.5942		
<u>block*cage</u>	<u>3</u>	<u>23.9475</u>	Error-a	
cultivar	2	88.4732	63.90	0.0001
cage*cultivar	2	14.6372	10.58	0.0022
<u>cultivar*block(cage)</u>	<u>12</u>	<u>8.3033</u>	Error-b	
time	1	1.2469	0.46	0.5061
cage*time	1	0.0003	0.00	0.9920
cultivar*time	2	7.5539	1.39	0.2735
cage*cultivar*time	2	8.3005	1.53	0.2430
<u>time*block(cage*cultivar)</u>	<u>18</u>	<u>48.7583</u>	Error-c	
Fungicide	2	6.9955	1.33	0.2699
cage*Fungicide	2	2.6756	0.51	0.6026
cultivar*Fungicide	4	7.1111	0.68	0.6095
time*Fungicide	2	1.4022	0.27	0.7662
cage*cultivar*Fungicide	4	6.6844	0.64	0.6373
cage*time*Fungicide	2	4.2022	0.80	0.4528
cultivar*time*Fungicide	4	4.4044	0.42	0.7938
cage*cultivar*time*Fungicide	4	3.6444	0.35	0.8450
<u>Fungi*block(cage*cvs*time)</u>	<u>72</u>	<u>188.8267</u>	Error-d	

Table.2

SUMMER 1981

General Linear Model Procedure

Dependent Variable: % Fruit set

<u>Sources</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>Mean-Square</u>		
Model	143	12600.0131	88.1119		
<u>Sources</u>	<u>DF</u>	<u>Type IV SS</u>	<u>F Value</u>	<u>Pr > F</u>	
cage	1	37.4136	0.11	0.7577	
block	3	111.6431			
block*cage	3	982.8075	Error-a		
cultivar	2	1476.7539	8.21	0.0057	
cage*cultivar	2	164.4506	0.91	0.4271	
cultivar*block (cage)	12	1079.4978	Error-b		
time	1	17.2225	0.30	0.5895	
cage*time	1	142.0069	2.49	0.1321	
cultivar*time	2	75.0050	0.66	0.5303	
cage*cultivar*time	2	68.5372	0.60	0.5591	
time*block (cage*cultivar)	18	1027.1550	Error-c		
Fungicide	2	184.2272	1.02	0.3665	
cage*Fungicide	2	105.1606	0.58	0.5619	
cultivar*Fungicide	4	260.0044	0.72	0.5822	
time*Fungicide	2	50.0217	0.28	0.7593	
cage*cultivar*Fungicide	4	54.5978	0.15	0.9620	
cage*time*Fungicide	2	1.2439	0.01	0.9932	
cultivar*time*Fungicide	4	218.0533	0.60	0.6621	
cage*cultivar*time*Fungicide	4	28.6444	0.08	0.9885	
Fungi*block (cage*cvs*time)	72	6515.5667	Error-d		

Table.3

SUMMER 1981

General Linear Model Procedure

Dependent Variable: Fruit Weight

<u>Sources</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>Mean-Square</u>		
Model	143	33312.1664	232.9522		
<u>Sources</u>	<u>DF</u>	<u>Type IV SS</u>	<u>F Value</u>	<u>Pr > F</u>	
cage	1	337.0069	1.33	0.3320	
block	3	374.8897			
block*cage	3	849.0542	Error-a		
cultivar	2	12625.12	134.17	0.0001	
cage*cultivar	2	2697.84	28.67	0.0001	
cultivar*block (cage)	12	564.5778	Error-b		
time	1	63.2025	0.51	0.4840	
cage*time	1	15.0802	0.12	0.7311	
cultivar*time	2	393.4516	1.59	0.2313	
cage*cultivar*time	2	136.8239	0.55	0.5848	
time*block (cage*cultivar)	18	2227.6217	Error-c		
Fungicide	2	447.4689	1.68	0.1928	
cage*Fungicide	2	172.2689	0.65	0.5259	
cultivar*Fungicide	4	792.1944	1.49	0.2140	
time*Fungicide	2	242.8067	0.91	0.4056	
cage*cultivar*Fungicide	4	615.4077	1.16	0.3365	
cage*time*Fungicide	2	136.7022	0.51	0.6000	
cultivar*time*Fungicide	4	535.1467	1.01	0.4096	
cage*cultivar*time*Fungicide	4	480.6511	0.90	0.4660	
Fungi*block (cage*cvs*time)	72	9564.8466	Error-d		

Table.4

SUMMER 1981

General Linear Model Procedure

Dependent Variable: Fruit Shape

<u>Sources</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>Mean-Square</u>		
Model	143	332.7724	2.3271		
<u>Sources</u>	<u>DF</u>	<u>Type IV SS</u>	<u>F Value</u>	<u>Pr > F</u>	
cage	1	2.0880	1.18	0.3575	
block	3	10.6138			
block*cage	3	5.3263	Error-a		
cultivar	2	117.8549	76.95	0.0001	
cage*cultivar	2	24.1128	15.74	0.0004	
cultivar*block(cage)	12	9.1895	Error-b		
time	1	6.8295	4.66	0.0446	
cage*time	1	0.0125	0.01	0.9275	
cultivar*time	2	1.3668	0.47	0.6345	
cage*cultivar*time	2	0.4803	0.16	0.8500	
time*block(cage*cultivar)	18	26.3627	Error-c		
Fungicide	2	4.3809	1.51	0.2273	
cage*Fungicide	2	0.3325	0.11	0.8917	
cultivar*Fungicide	4	5.7512	0.99	0.4171	
time*Fungicide	2	0.7538	0.26	0.7716	
cage*cultivar*Fungicide	4	2.0212	0.35	0.8439	
cage*time*Fungicide	2	0.1063	0.04	0.9460	
cultivar*time*Fungicide	4	5.9895	1.03	0.3957	
cage*cultivar*time*Fungicide	4	4.9357	0.85	0.4970	
Fungi*block(cage*cvs*time)	72	104.2644	Error-d		

Table.5

SUMMER 1981

General Linear Model Procedure

Dependent Variable: Fruit Disease

<u>Sources</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>Mean-Square</u>
Model	143	91.4123	0.6392

<u>Sources</u>	<u>DF</u>	<u>Type IV SS</u>	<u>F Value</u>	<u>Pr > F</u>
cage	1	6.4516	31.52	0.0001
block	3	0.3393		
<u>block*cage</u>	<u>3</u>	<u>0.6140</u>	Error-a	
cultivar	2	24.6100	27.52	0.0001
cage*cultivar	2	2.3777	2.66	0.1107
<u>cultivar*block(cage)</u>	<u>12</u>	<u>5.3651</u>	Error-b	
time	1	1.5211	4.31	0.0524
cage*time	1	0.2756	0.78	0.3883
cultivar*time	2	2.9528	4.19	0.0321
cage*cultivar*time	2	0.1651	0.23	0.7937
<u>time*block(cage*cultivar)</u>	<u>18</u>	<u>6.3483</u>	Error-c	
Fungicide	2	2.2074	2.66	0.0769
cage*Fungicide	2	0.2795	0.34	0.7152
cultivar*Fungicide	4	2.6593	1.60	0.1832
time*Fungicide	2	0.0869	0.10	0.9007
cage*cultivar*Fungicide	4	2.7531	1.66	0.1691
cage*time*Fungicide	2	0.0621	0.07	0.9280
cultivar*time*Fungicide	4	1.7989	1.08	0.3711
cage*cultivar*time*Fungicide	4	0.6613	0.40	0.8092
<u>Fungi*block(cage*cvs*time)</u>	<u>72</u>	<u>29.8831</u>	Error-d	

Table.6

SUMMER 1981

General Linear Model Procedure

Dependent Variable: Log10 % Fruit Set

<u>Sources</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>Mean-Square</u>
Model	143	1.2318	0.0086

<u>Sources</u>	<u>DF</u>	<u>Type IV SS</u>	<u>F Value</u>	<u>Pr > F</u>
cage	1	0.0136	1.14	0.3639
block	3	0.0246		
<u>block*cage</u>	<u>3</u>	<u>0.0562</u>	Error-a	
cultivar	2	0.0588	4.81	0.0292
cage*cultivar	2	0.0000	00.00	0.9978
<u>cultivar*block(cage)</u>	<u>12</u>	<u>0.0732</u>	Error-b	
time	1	0.0001	0.01	0.9075
cage*time	1	0.0053	0.55	0.4685
cultivar*time	2	0.0099	0.50	0.6131
cage*cultivar*time	2	0.0007	0.04	0.9625
<u>time*block(cage*cultivar)</u>	<u>18</u>	<u>0.1765</u>	Error-c	
Fungicide	2	0.0311	1.66	0.1967
cage*Fungicide	2	0.0053	0.28	0.7534
cultivar*Fungicide	4	0.0077	0.21	0.9339
time*Fungicide	2	0.0016	0.09	0.9160
cage*cultivar*Fungicide	4	0.0193	0.52	0.7224
cage*time*Fungicide	2	0.0013	0.07	0.9320
cultivar*time*Fungicide	4	0.0458	1.22	0.3085
cage*cultivar*time*Fungicide	4	0.0271	0.72	0.5789
<u>Fungi*block(cage*cvs*time)</u>	<u>72</u>	<u>0.6733</u>	Error-d	

Table.7

SUMMER 1981**General Linear Model Procedure**Dependent Variable: Total of Sound Achenes

<u>Sources</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
Model	71	1142722.1015	4.37	0.0001	0.8117	14.2
				<u>Std.Dev.</u>		<u>Mean</u>
Total	143	1407794.2800		60.6758		428.16

<u>Sources</u>	<u>DF</u>	<u>ANOVA SS</u>	<u>F Value</u>	<u>Pr > F</u>
cage	1	49989.1343	20.96	0.0196
block	3	52578.6711	4.76	0.0045
<u>block*cage</u>	<u>3</u>	<u>7154.9920</u>	Error-a	
cultivar	2	777459.0604	4.81	0.0292
cage*cultivar	2	3495.4875	0.70	0.5166
<u>cultivar*block(cage)</u>	<u>12</u>	<u>30034.5567</u>	Error-b	
time	1	27954.4467	5.96	0.0251
cage*time	1	58.3569	0.01	0.9124
cultivar*time	2	7902.3557	0.84	0.9124
cage*cultivar*time	2	197.9076	0.02	0.9791
<u>time*block(cage*cultivar)</u>	<u>18</u>	<u>83368.1892</u>	Error-c	
Fungicide	2	17645.9308	2.40	0.0983
cage*Fungicide	2	10116.4457	1.37	0.2597
cultivar*Fungicide	4	16253.6801	1.10	0.3615
time*Fungicide	2	3251.7166	0.44	0.6447
cage*cultivar*Fungicide	4	7399.0458	0.50	0.7340
cage*time*Fungicide	2	20954.2243	2.85	0.0646
cultivar*time*Fungicide	4	17739.8333	1.20	0.3163
cage*cultivar*time*Fungicide	4	8168.0668	0.55	0.6962
<u>Fungi*block(cage*cvs*time)</u>	<u>72</u>	<u>265072.1786</u>	Error-d	

Table.8

GREENHOUSE EXPERIMENT

Analysis of Variance Procedure

Dependent Variable: Yield

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
model	23	1280.37	24.46	0.0001	0.9036	19.5
error	60	136.57		Stn Dev	Mean	
total	83	1416.94		1.5087	7.7372	

<u>Source</u>	<u>DF</u>	<u>ANOVA SS</u>	<u>F Value</u>	<u>Pr > F</u>
block	3	2.3417	0.34	0.7967
cultivar	2	29.8048	6.55	0.0027
treatment	6	1154.5168	84.53	0.0001
cvs*trt	12	93.7066	3.43	0.0007

Table.9

GREENHOUSE EXPERIMENT

Analysis of Variance Procedure

Dependent Variable: Shape

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
model	23	180.44	36.76	0.0001	0.9337	15.8
error	60	12.80		Stn Dev	Mean	
total	83	193.25		0.4619	2.9226	

<u>Source</u>	<u>DF</u>	<u>ANOVA SS</u>	<u>F Value</u>	<u>Pr > F</u>
block	3	1.0089	1.58	0.2032
cultivar	2	6.3631	14.91	0.0001
treatment	6	160.3512	125.24	0.0001
cvs*trt	12	12.7202	4.97	0.0001

Table.10

GREENHOUSE EXPERIMENT

Analysis of Variance Procedure

Dependent Variable: Total achenes

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
model	23	69997.7649	3.53	0.0001	0.5751	14.1

error	60	5172.4107		Stn Dev	Mean
total	83	121720.1756		29.3605	208.4702

<u>Source</u>	<u>DF</u>	<u>ANOVA SS</u>	<u>F Value</u>	<u>Pr > F</u>
block	3	3370.6518	1.30	0.2811
cultivar	2	2001.2381	1.16	0.3202
treatment	6	41201.3631	7.97	0.0001
cvs*trt	12	23424.5119	2.26	0.0192

Table.11

GREENHOUSE EXPERIMENT

Analysis of Variance Procedure

Dependent Variable: Aborted Achenes

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
model	23	256824.8631	16.57	0.0001	0.8640	35.1

error	60	40437.1250		Stn Dev	Mean
total	83	297261.9881		25.9606	73.9881

<u>Source</u>	<u>DF</u>	<u>ANOVA SS</u>	<u>F Value</u>	<u>Pr > F</u>
block	3	2231.7500	1.10	0.3552
cultivar	2	4682.8988	3.47	0.0374
treatment	6	227934.1964	56.37	0.0001
cvs*trt	12	21976.0179	2.72	0.0054

Table.12

GREENHOUSE EXPERIMENT

Analysis of Variance Procedure

Dependent Variable: Sound Achenes

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr>F</u>	<u>R-Square</u>	<u>C.V.</u>
model	23	427621.6584	19.69	0.0001	0.8830	22.8
error	60	56647.1547		Stn Dev	Mean	
total	83	484268.7232		30.7265	134.4821	

<u>Source</u>	<u>DF</u>	<u>ANOVA SS</u>	<u>F Value</u>	<u>Pr > F</u>
block	3	1596.0327	0.56	0.6451
cultivar	2	12806.7321	6.78	0.0022
treatment	6	393311.1607	69.43	0.0001
cvs*trt	12	19907.6428	1.76	0.0769

Table.13

GREENHOUSE EXPERIMENT

Analysis of Variance Procedure

Dependent Variable: Aborted Achenes "SQRT"

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr>F</u>	<u>R-Square</u>	<u>C.V.</u>
model	23	519.8547	19.34	0.0001	0.8811	19.4
error	60	70.1263		Stn Dev	Mean	
total	83	589.9810		1.0811	5.5653	

<u>Source</u>	<u>DF</u>	<u>ANOVA SS</u>	<u>F Value</u>	<u>Pr > F</u>
block	3	1.0691	0.30	0.8232
cultivar	2	20.9838	8.98	0.0004
treatment	6	474.6444	67.68	0.0001
cvs*trt	12	23.1570	1.65	0.1017

Table.14

GREENHOUSE EXPERIMENT
Analysis of Variance Procedure

Dependent Variable: Sound Achenes "SQRT"

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
model	23	796.8210	64.16	0.0001	0.9609	10.2

error	60	32.39		Stn Dev	Mean
total	83	329.22		0.7348	7.2203

<u>Source</u>	<u>DF</u>	<u>ANOVA SS</u>	<u>F Value</u>	<u>Pr > F</u>
block	3	1.6283	1.01	0.3980
cultivar	2	5.3713	4.97	0.0100
treatment	6	790.1116	240.79	0.0001
cvs* trt	12	9.7097	1.50	0.1501

Table.15

Winter 1982Laboratory ExperimentDependent Variable: % Pollen Germination

<u>Source</u>	<u>DF</u>	<u>SS</u>	<u>F value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
Model	125	860.78	31.51	0.0001	0.9690	21.1
Error-b	126	27.54				
Total	251	888.31		Std Dev		Mean
				0.4675		2.22

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>
cultivar	2	1.31	3.00	0.5533
Treat	6	764.99	583.42	0.0001
cultivar*Treat	12	5.47	2.09	0.9482
Rep(cultivar*Treat)	42	45.89	Error-a	
Time	3	6.32	9.64	0.0001
Treat*Time	18	18.47	4.69	0.0001
cultivar*Time	6	5.82	4.44	0.0004
cultivar*Treat*Time	36	12.51	1.59	0.0322

Table.16

Winter 1982Laboratory ExperimentDependent Variable: length of pollen tube

<u>Source</u>	<u>DF</u>	<u>SS</u>	<u>F value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
Model	125	1.0951	17.71	0.0001	0.9461	3.0
Error-b	126	0.0623				
Total	251	1.1574		Std Dev		Mean
				0.0222		0.7388

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>
cultivar	2	0.0060	6.04	0.0836
Treat	6	0.7347	247.55	0.0001
cultivar*Treat	12	0.0092	1.56	0.7618
Rep(cultivar*Treat)	42	0.0476	Error-a	
Time	3	0.0739	49.15	0.0001
Treat*Time	18	0.2044	22.95	0.0001
cultivar*Time	6	0.0048	1.61	0.1503
cultivar*Treat*Time	36	0.0155	0.87	0.6772

Table.17

Summer 1982Analysis of Variance ProcedureDependent Variable: Fruit Weight

<u>Source</u>	<u>DF</u>	<u>SS</u>	<u>F value</u>	<u>pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
<u>Model</u>	<u>17</u>	<u>13.59</u>	<u>4.71</u>	<u>0.0010</u>	<u>0.8165</u>	<u>25.9</u>
<u>Error-b</u>	<u>18</u>	<u>3.05</u>		<u>Std.Dev.</u>		<u>Mean</u>
<u>Total</u>	<u>35</u>	<u>16.64</u>		<u>0.4118</u>		<u>1.5848</u>

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>
<u>Block</u>	<u>3</u>	<u>0.8417</u>		
<u>cultivar</u>	<u>2</u>	<u>10.4681</u>	<u>44.38</u>	<u>0.0003</u>
<u>Block*cultivar</u>	<u>6</u>	<u>0.7076</u>	<u>Error-a</u>	
<u>Fungicide</u>	<u>2</u>	<u>0.1026</u>	<u>0.30</u>	<u>0.7427</u>
<u>cultivar*Fungicide</u>	<u>4</u>	<u>1.4685</u>	<u>2.16</u>	<u>0.1145</u>

Table.18

SUMMER 1982Analysis of Variance ProcedureDependent Variable: Fruit Number

<u>Source</u>	<u>DF</u>	<u>SS</u>	<u>F Value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
<u>Model</u>	<u>17</u>	<u>269450.14</u>	<u>7.13</u>	<u>0.0001</u>	<u>0.8706</u>	<u>18.11</u>
<u>Error-b</u>	<u>18</u>	<u>40034.17</u>		<u>Std.Dev.</u>		<u>Mean</u>
<u>Total</u>	<u>35</u>	<u>309484.31</u>		<u>47.16</u>		<u>260.36</u>

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>
<u>Block</u>	<u>3</u>	<u>18354.97</u>		
<u>cultivar</u>	<u>2</u>	<u>202211.72</u>	<u>21.98</u>	<u>0.0017</u>
<u>Block*cultivar</u>	<u>6</u>	<u>27595.61</u>	<u>Error-a</u>	
<u>Fungicide</u>	<u>2</u>	<u>1320.06</u>	<u>0.30</u>	<u>0.7468</u>
<u>cultivar*Fungicide</u>	<u>4</u>	<u>19967.78</u>	<u>2.24</u>	<u>0.1047</u>

Table.19

SUMMER 1982

Analysis of Variance Procedure

Dependent Variable: Fruit Shape

<u>Source</u>	<u>DF</u>	<u>SS</u>	<u>F value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
Model	17	12.13	10.02	0.0001	0.9044	10.0
Error-b	18	1.28				
Total	35	13.41				
			Std.Dev.		Mean	
			0.2668		2.6828	

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>
Block	3	0.7164		
cultivar	2	9.8422	58.53	0.0001
Block*cultivar	6	0.5044	Error-a	
Fungicide	2	0.5739	4.03	0.0358
cultivar*Fungicide	4	0.4911	1.72	0.1885

Table.20

SUMMER 1982

Analysis of Variance Procedure

Dependent Variable: Fruit Disease

<u>Source</u>	<u>DF</u>	<u>SS</u>	<u>F Value</u>	<u>Pr > F</u>	<u>R-Square</u>
Model	17	32.8056	1.52	0.1927	0.5862
Error-b	18	22.8333			
Total	35	55.6389			
			Std.Dev.		Mean
			1.1263		0.8056

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>
Block	3	1.1944		
cultivar	2	1.7222	0.48	0.6396
Block*cultivar	6	10.7222	Error-a	
Fungicide	2	16.7222	6.59	0.0071
cultivar*Fungicide	4	2.4444	0.48	0.7488

Table.21

SUMMER 1982

Analysis of Variance Procedure

Dependent Variable: Total Achenes

<u>Source</u>	<u>DF</u>	<u>SS</u>	<u>F value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
Model	17	140351.67	1.59	0.1695	0.6000	20.6
Error-b	18	93562.33				
Total	35	233914.00				
			<u>Std.Dev.</u>		<u>Mean</u>	
			72.0965		349.7	

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>
Block	3	25784.22		
cultivar	2	19430.17	0.87	0.4663
Block*cultivar	6	67092.94	Error-a	
Fungicide	2	12205.50	1.17	0.3317
cultivar*Fungicide	4	15838.83	0.76	0.5636

Table.22

SUMMER 1982

Analysis of Variance Procedure

Dependent Variable: Aborted Achenes

<u>Source</u>	<u>DF</u>	<u>SS</u>	<u>F Value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
Model	17	55744.47	2.05	0.0704	0.6593	43.98
Error-b	18	28816.50				
Total	35	84590.97				
			<u>Std.Dev.</u>		<u>Mean</u>	
			40.01		90.92	

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>
Block	3	2850.75		
cultivar	2	12128.39	1.80	0.2434
Block*cultivar	6	20158.50	Error-a	
Fungicide	2	11024.06	3.44	0.0542
cultivar*Fungicide	4	9612.78	1.50	0.2437

Table.23

SUMMER 1982

Analysis of Variance Procedure

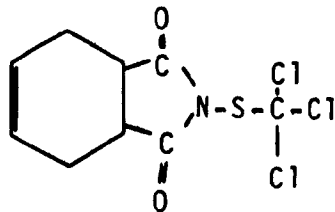
Dependent Variable: Sound Achenes

<u>Source</u>	<u>DF</u>	<u>SS</u>	<u>F Value</u>	<u>Pr > F</u>	<u>R-Square</u>	<u>C.V.</u>
Model	17	96172.81	1.13	0.3993	0.5161	27.36
Error-b	18	90172.83				
Total	35	186345.64				
			<u>Std.Dev.</u>		<u>Mean</u>	
			70.78		258.69	

<u>Source</u>	<u>DF</u>	<u>Sum of Squares</u>	<u>F Value</u>	<u>Pr > F</u>
Block	3	13084.97		
cultivar	2	24682.06	1.71	0.2580
Block*cultivar	6	43423.94	Error-a	
Fungicide	2	10137.56	1.01	0.3833
cultivar*Fungicide	4	5024.28	0.25	0.9054

APPENDIX 2
DESCRIPTION OF CHEMICALS

Trade Names: Captan, Orthocide 406, Vancide 89



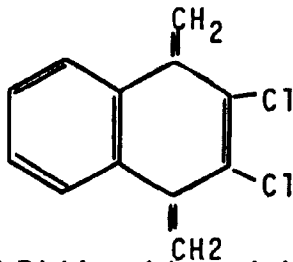
N- (trichloromethylthio)-4-cyclohexane-1,2-dicarboximide

Type: is a chlorinated hydrocarbon used as a protective fungicide.

Toxicity: LD₅₀; 9000 mg/kg

available Formulations: 50% WP, dusts 5, 7.5, 10 and 20%; 75% powder for seed treatment.

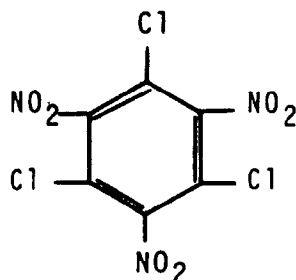
Trade Names: Dichlone, Phygon XL.



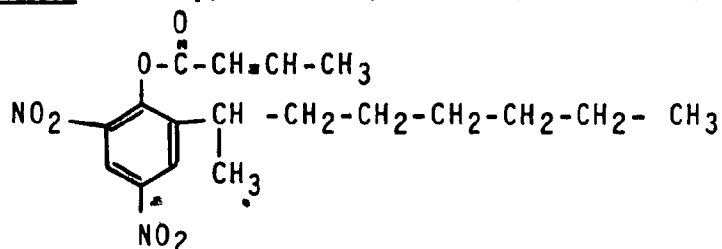
2,3-Dichloro-1,4- naphthoquinone

Type: Phygon is a chlorinated hydrocarbon used as a foliar and soil fungicide as well as a seed treatment.

Toxicity: LD₅₀; 1300mg/kg, may be irritating to the skin.

Trade Names Bulbosan

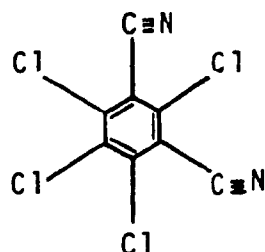
Trichlorotrinitrobenzene

Type: is an organic compound used as a foliar fungicide.Toxicity: LD₅₀; 10,000 mg/kg.available Formulations: 5% dusts TCNB: 1,2,4,5-tetrachloro-3-nitrobenzene- used to a limited extent as a fungicide in Europe.Trade Names Dinocap, Karathane, Arathane, Mildex, Capryl

2,4- dinitro-6-(2-octyl) phenyl crotonate

Type: is a dinitro compound used as a protective fungicide-acaricideToxicity: LD₅₀; 980 mg/kg, may cause skin irritationavailable Formulations: 1.5 and 10% dusts, 25% WP

Trade Names Chlorothalonil , Daconil 2787, Nopocide, termil, Exotherm, Bravo, Blazon



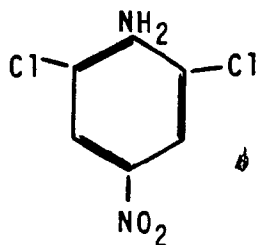
Tetrachloroisophthalonitrile

Types is an organic compound used as a preventative, foliar fungicide

Toxicity LD₅₀; 10,000 mg/kg

available Formulations: 75% WP, 90% tablets, 20% exothermic powder

Trade Names Dicloran, Botran, Allisan, DCNA



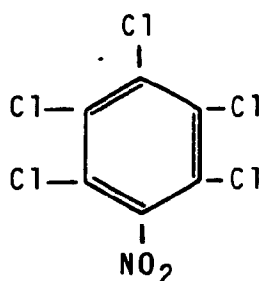
2,6- dichloro-4-nitroaniline

Types is a chlorinated hydrocarbon used as a soil and foliar fungicide.

Toxicity LD₅₀; 2000 mg/kg

available Formulations: 50%, 75% WP, 4 and 6% dusts.

Trade Names PCNB, Terrachlor, Brassicol, Tritisan, Folsosan, Botrylex, Tilcarex Fungiclor, Quintozene

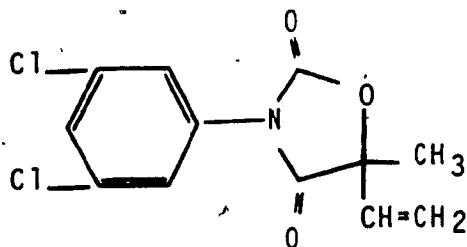


Pentachloronitrobenzene

Type: is a chlorinated hydrocarbon compound used as a soil fungicide and seed disinfectant

Toxicity: LD₅₀; 1650 mg/kg, may cause skin irritation

Trade Names Rovilan, Vinclozolin, BAS-3520, Ornalin



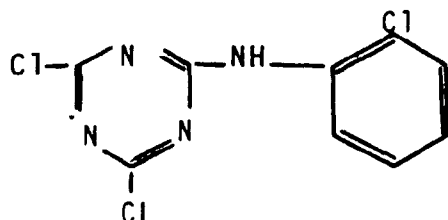
3-(3,5-dichlorophenyl)-5-ethynyl-5-methyl-2,4-oxazolidinedione

Type: Vinclozoline is an organic compound and protectant fungicide

Toxicity: LD₅₀; 10,000 mg/kg

available Formulations 50% WP

Trade Names Dyrene, Triazine, Kemate



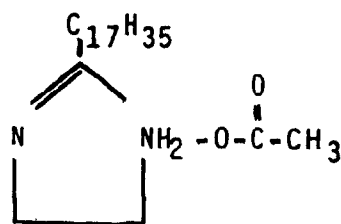
2,4,-dichloro-6-O-chloronilnotinotriazine

Type: is a triazine compound used as a foliar fungicide

Toxicity: LD₅₀; 2710 mg/kg

available Formulations: 50% WP, 5% dusts, granules 5%, also mixed with Dexon as a 50-50% WP

Trade Names Glyodin, Glyoxide, Crag fruit fungicide 341, Glyoxalidine



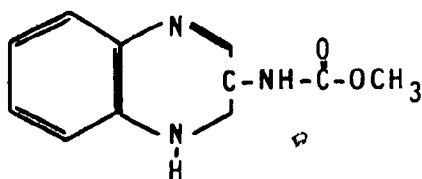
2-heptadecyl-imidazoline 2-heptadecylglyoxalidine acetate

Type: is an organic compound used as a protective foliar fungicide

Toxicity: LD₅₀; 372 mg/kg

available Formulations: 70% WP, 30% solution

Trade Names Carbendazin, MBC, Bavistin, Derrosal, Hoe 17411 (Hoechst AG), Kemdazon.



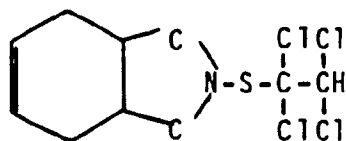
2-(Methoxycarbonylamino)-benzimidazole

Type Systemic fungicide

Toxicity LD₅₀; 15,000 mg/kg

Available Formulations 50% 60% WP

Trade Names Captafol, Difolatan, Sulfenimide, Folcid



N-(1,1,2,2-tetrachloroethylsulfonyl)-cis-Δ⁴-cyclohexene-1,2-dicarboximide.

Type Sulfenimide is an organic chlorinated hydrocarbon used as a foliar fungicide.

Toxicity LD₅₀; 4600 mg/kg

available Formulations 80% WP, 5 and 7.5% dusts.

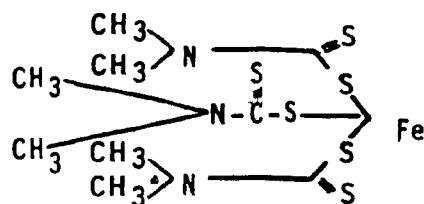
Trade Names Dithane M-45, Mancozeb (150-Europe), Manzeb, Manzin 80, Nemispor MZ, Policar S, Ziman-dithane

Maneb + Zinc ion

Chemical Composition A ready-to-use agricultural fungicide which contains 80% active ingredient. It is a coordination product of zinc ion and manganese ethylene bisdithiocarbamate and is related to both Manzeb and Zineb.

available Formulations 80% N and dust

Trade Names Ferbam, Fermate, Vancide F, Carbamate, Coromate, Karbam black, Fermocide, Ferberk, Ferradow, Niacide, Fermulene, Fermate-D, Trifungol



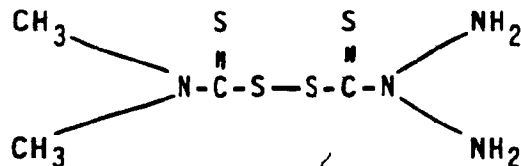
Ferric dimethyldithiocarbamate

Type: is a carbamate compound used as a foliar fungicide

Toxicity: LD₅₀; 1000 mg/kg

available Formulations: 76% WP, 3.9, 7.6 and 11.5% dusts

Trade Names Thiram, Tripomol, TMTD, TMTDS, Thylate, Pomarsolforate, Fernacol, Arasan 75, Nomersan, Pomasol, Spotrate, Thiramade, Turf-tox MC, Ranoran 75, Thiosan, Fernasan, Nangets



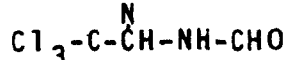
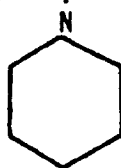
Tetramethylthiuramdisulfide

Type is a organic compound use as a protective foliage fungicide, seed treatment and animal repellent

Toxicity LD₅₀ ; 865 mg/kg

available Formulations 40% 50% WP

Trade Names Triforine, Cela W-524, CME 74770, Funginex, saprol

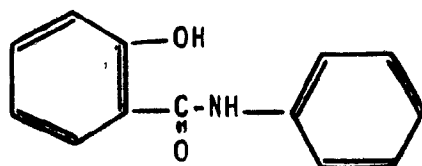


N-N'-(1,4-piperazinediyl-BIS(2,2,2-trichloroethylidene))- BIS-(formamide)

Type Locally systemic fungicide (protectant, eradicant and curative) fungicide.

Toxicity LD₅₀ ; 16000 mg/kg

available Formulations EC (emulsifiable concentrate) 18.2% by weight

Trade Names Salicylanilide, Shirlan**Salicylanilide**

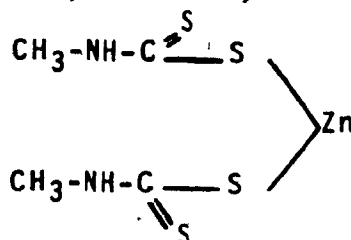
Types is an organic compound shown to be both a fungicide and a bactericide

Toxicity LD₅₀; 2000 mg/kg

available Formulations various

Zinc salicylanilides This compound, mixed with colloidal sulfur, is used in Russia as a plant fungicide.

Trade Names Zeneb, Dithane Z-78, Parzate C zineb, Aspor, Tiezene, Lonacol, Zimate, Partoftoral, Zenbenide, Blitex

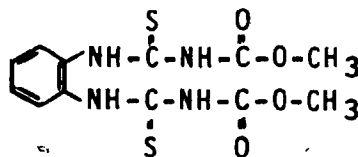
**Zinc ethylene BIS dithiocarbamate**

Types is a carbamate compound used as protective foliar fungicide.

Toxicity LD₅₀; 5200 mg/kg, may cause irritation to the nose.

available Formulations 65%, 70%, 75%, 80% WP, dusts 3.9, 4.6, 7.6, 10 and 16.25%

Trade Names Easout, Thiophanate methyl, Cercobin methyl, Topsin WP methyl, Mildothane, Trevin, Fungo, Topsin-M, Cercobin-M, Enovit-Super, Pelt-44, Cycosin, Dousan, Dinorsan, Sigma, Labilite



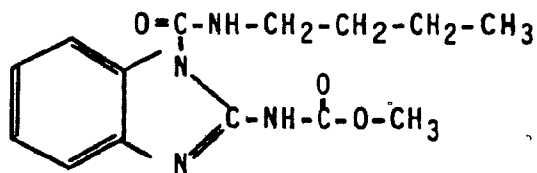
Dimethyl 4,4'-O-phenylenebis (3-thioallophanate)

Types is an organic compound used as a curative, preventive and systemic fungicide.

Toxicity LD₅₀; 7500 mg/kg

available Formulations 50% 70% WP

Trade Names Benomyl (ASA, BSI, ISO), Benlate, Tersan 1991



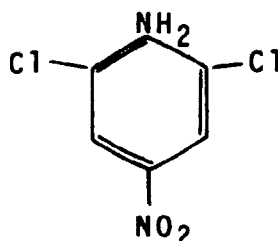
Methyl 1-(butylcarbamoyl)-1H-benzimidazolecarbamate

Types Systemic fungicide

Toxicity LD₅₀; 10,000 mg/kg

available Formulations 50% WP

Trade Names Botran, Dicloran, Allisan, DCNA



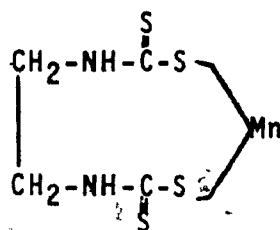
2,6-dichloro-4-nitroaniline

Type is a chlorinated hydrocarbon used as a soil and foliar fungicide.

Toxicity LD₅₀; 2000 mg/kg.

available Formulations 50% 75% WP, 4 and 6% dusts

Trade Names Maneb, MEB, MNEBD, Manzate, Dithane M-22, Vancid M, Manzate-D, Lonocol M, Dithane, Trimangol



Manganous ethylene BIS dithiocarbamate

Type is a carbamate compound used as a foliar fungicide.

Toxicity LD₅₀; 6750 mg/kg.

available Formulations 70%, 80% WP, dusts 3.5, 5.6, 6, 7 and 8%

APPENDIX 3
SOIL ANALYSIS PROCEDURE

Table.1

SOIL TEST REPORTName: Khanizadeh, Shahrokh Date Sampled: May, 15, 1981Sample No: _____ Date tested: June, 10, 1981

Description of Location and Topography: Located on hill, with slight slope toward south West and windy

Texture: Clay loam Color _____ Drainage: Fairly good

Past Management (Last Crop, Treatment, etc.)

Melon, Tomato & Eggplant

	Results	Comments
pH	6.6-6.2	
Nitrate Nitrogen	10lb/acre	
Phosphorus(P)(Px2.3=P2o5)	200lb/acre	
Potassium (K)(Kx1.2=K2o)	350lb/acre	
Humus	High: consider as agricultural soil	
Calcium		
Ammonia Nitrogen	very low	
Magnesium	low	
Manganese	medium low-low	
Aluminum (active)	very low	
Nitrate Nitrogen	1 ppm: even less than this	
Ferric Iron	5lb/acre	
Sulfate	50 ppm	
Chloride	100 ppm	

NOTE: $\text{lb acre}^{-1/2} = \text{ppm}$ and $(\text{ppm}) \times 2 = \text{lb acre}^{-1}$

PROCEDURE FOR PARTICLE SIZE ANALYSIS

In order to analyze the particle size of the soil, the following procedure was carried out.

- 1- Fifty grams of field soil was weighed into dry, 250 ml beaker.
- 2- Enough distilled H_2O (approx. 150 ml) was added to cover the soil.
- 3- Twenty five ml of Sodium Hexametaphosphate was added to the solution from a burette.
- 4- The suspension was stirred intermittently with a stirring rod for 15 mins.
- 5- The suspension was poured into a mixing cup, making sure that the soil particles on the sides of the beaker were washed off with a washbottle into the cup. Extra water was added in order to half fill the cup.
- 6- The suspension was mixed for five minutes using an electric mixer.
- 7- The contents of the cup was poured into a one litre cylinder. Again, care was taken to wash the contents of the soil into the cylinder. The cylinder was filled to the 1000 ml mark with H_2O .
- 8- When the time came to take a reading, the soil suspension was thoroughly mixed by plunging it vigorously 10 times with the metal plunger.

After the stirring was completed, the suspension was transferred

to a special cylinder, with the aid of a wash bottle. Distilled water was added to the cylinder, to bring the volume to exactly 1,000 ml. A stirring plunger was placed in the cylinder and the plunger was pushed down and pulled up several times to get all the soil particles in suspension. When this was accomplished, the plunger was removed and the time noted immediately. Any foam that formed on the surface was quickly dissipated by the addition of 1 or 2 drops of amylalcohol. About 20 seconds before the reading was to be taken, a hydrometer was carefully inserted. After obtaining the reading, the hydrometer was carefully removed. The hydrometer was rinsed and set aside until the next reading was to be taken.

Since the viscosity of a liquid medium changes with temperature, it was necessary to record the temperature of the suspension at each reading. For each degree above 20°C , 0.36 g of sample was added to get the corrected hydrometer reading. For each degree below 20°C , 0.36 g was subtracted.

The results that were obtained are tabulated in the next 3 pages.

NOTE:

from: Soil Science Department, Macdonald Campus of McGill University

Sample 1.

Particle Size Analysis

	top soil 5-10cm	soil 10-20cm
1- Sample size (g)	50	50
2- 40 Sec. hydrometer reading g/l	25	29
3- 40 Sec. temp. C	26	26
4- 40 Sec. hydrometer reading corrected g/l	27.16	31.16
5- 2 hr. hydrometer reading g/l	13	13
6- 2 hr. temp. C	25	26
7- 2 hr. hydrometer reading corrected	14.8	15.16
8- per cent sand= $(1-4)/1 * 100$	45.68%	37.68%
9- per cent clay= $7/1 * 100$	29.60%	30.32%
10- per cent silt= $100 - (8+9)$	24.72%	32.00%
11- textural class		
12- per cent rock (wt. of rocks) $*100/50=$	05.20%	03.12%

Note:

Sand bigger than 2 mm is considered to be rocks

Sample 2.

Particle Size Analysis

	top soil 5-10cm	soil 10-20cm
1- Sample size (g)	50	50
2- 40 Sec. hydrometer reading g/l	28	33
3- 40 Sec. temp. C	26	26
4- 40 Sec. hydrometer reading corrected g/l	30.16	35.16
5- 2 hr. hydrometer reading g/l	15	15
6- 2 hr. temp. C	25.5	26
7- 2 hr. hydrometer reading corrected	16.98	17.16
8- per cent sand= $(1-4)/1 * 100$	39.68%	29.68%
9- per cent clay= $7/1 * 100$	33.96%	34.32%
10- per cent silt= $100 - (8+9)$	26.36%	36.00%
11- textural class		
12- per cent rock (wt. of rocks) $*100/50=$. . .	02.00%	00.60%

Note:

Sand bigger than 2 mm is considered to be rocks

Sample 3

Particle Size Analysis

	top soil 5-10cm	soil 10-20cm
1- Sample size (g)	50	50
2- 40 Sec. hydrometer reading g/l	30	34
3- 40 Sec. temp. C	26	25.5
4- 40 Sec. hydrometer reading corrected g/l	32.16	36.16
5- 2 hr. hydrometer reading g/l	14	18
6- 2 hr. temp. C	26	25.5
7- 2 hr. hydrometer reading corrected	16.16	20.16
8- per cent sand= (1-4)/1 * 100	35.68%	27.68%
9- per cent clay= 7/1 * 100	32.32%	40.32%
10- per cent silt= 100 - (8+9)	32.00%	32.00%
11- textural class		
12- per cent rock (wt. of rocks) *100/50=	01.50%	00.60%

Note:

Sand bigger than 2 mm is considered to be rocks

APPENDIX 4

FIGURES

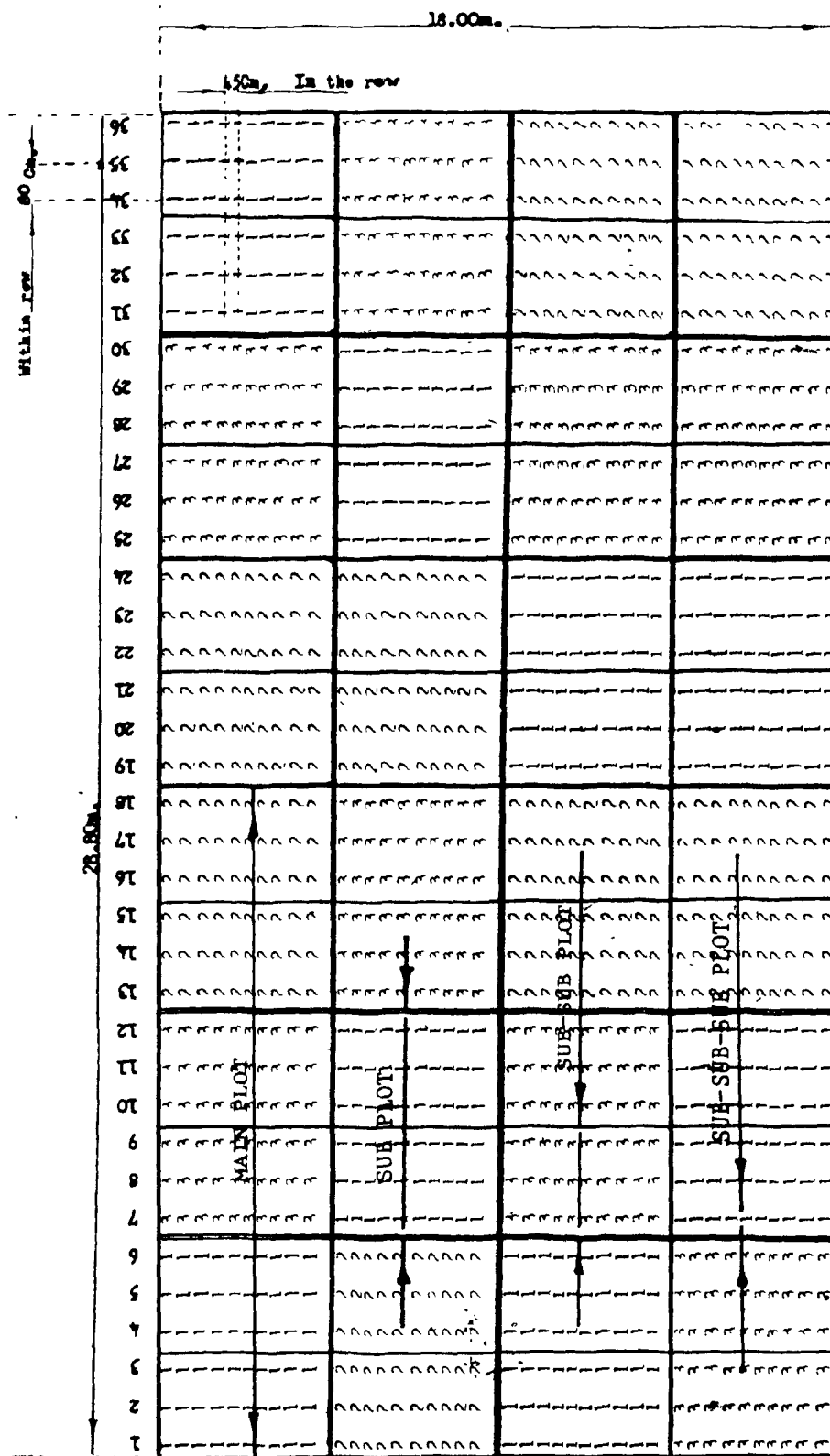


Figure 1. A schematic view of split plots using a randomized complete block design.

Note: Each no. in the field layout stands for one plant and different nos. indicate different varieties.

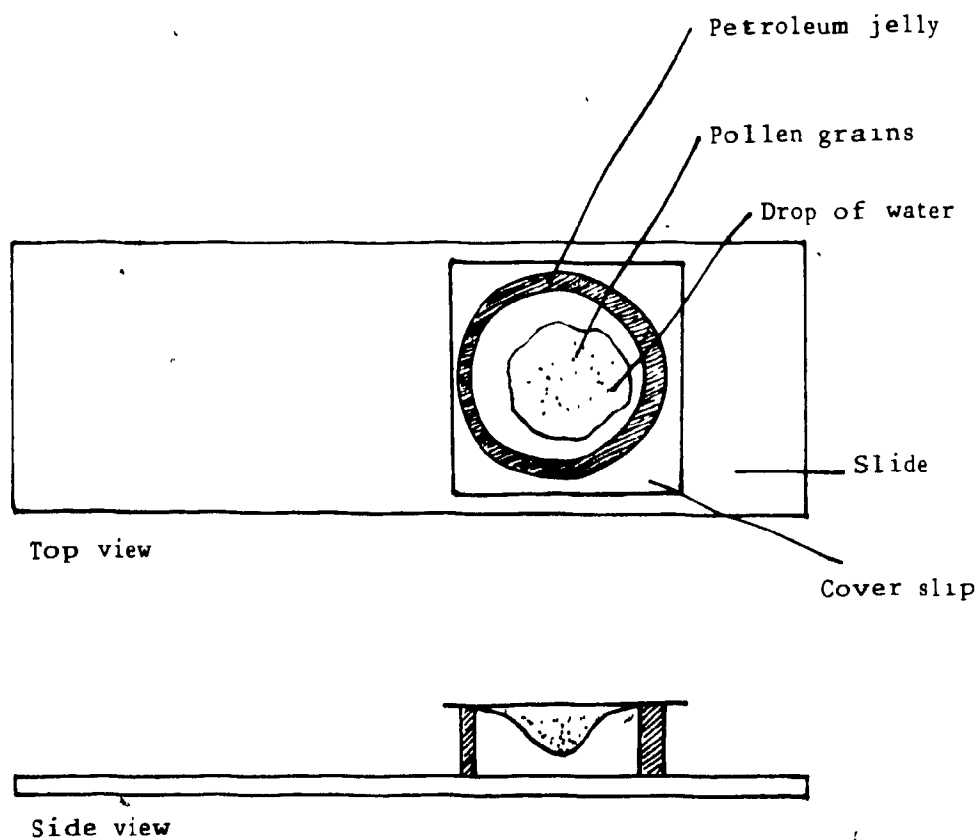


Figure 2. Hanging drop method used to count the percentage of pollen germination (Van Tieghem cell adapted for pollen tube culture).

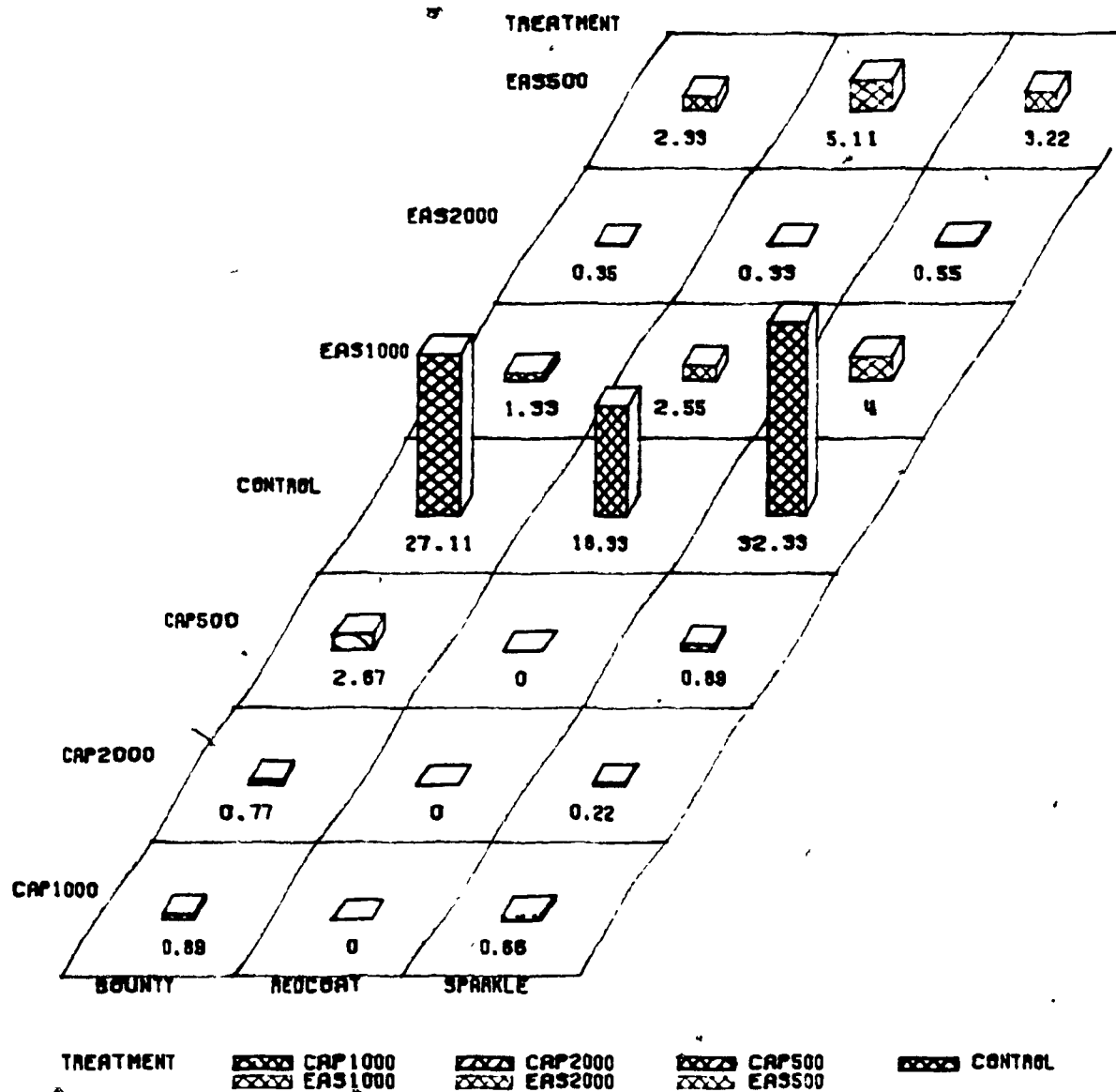


Figure 3. Percent pollen germination (after three hours) of three strawberry cvs. treated with different concentrations of fungicide in the germination media. Each bar represents the mean of three observations. EAS500 = Easout 500 ppm; EAS1000 = Easout 1000 ppm; EAS2000 = Easout 2000 ppm; CAP500 = Captan 500 ppm; CAP1000 = Captan 1000 ppm; CAP 2000 = Captan 2000 ppm.

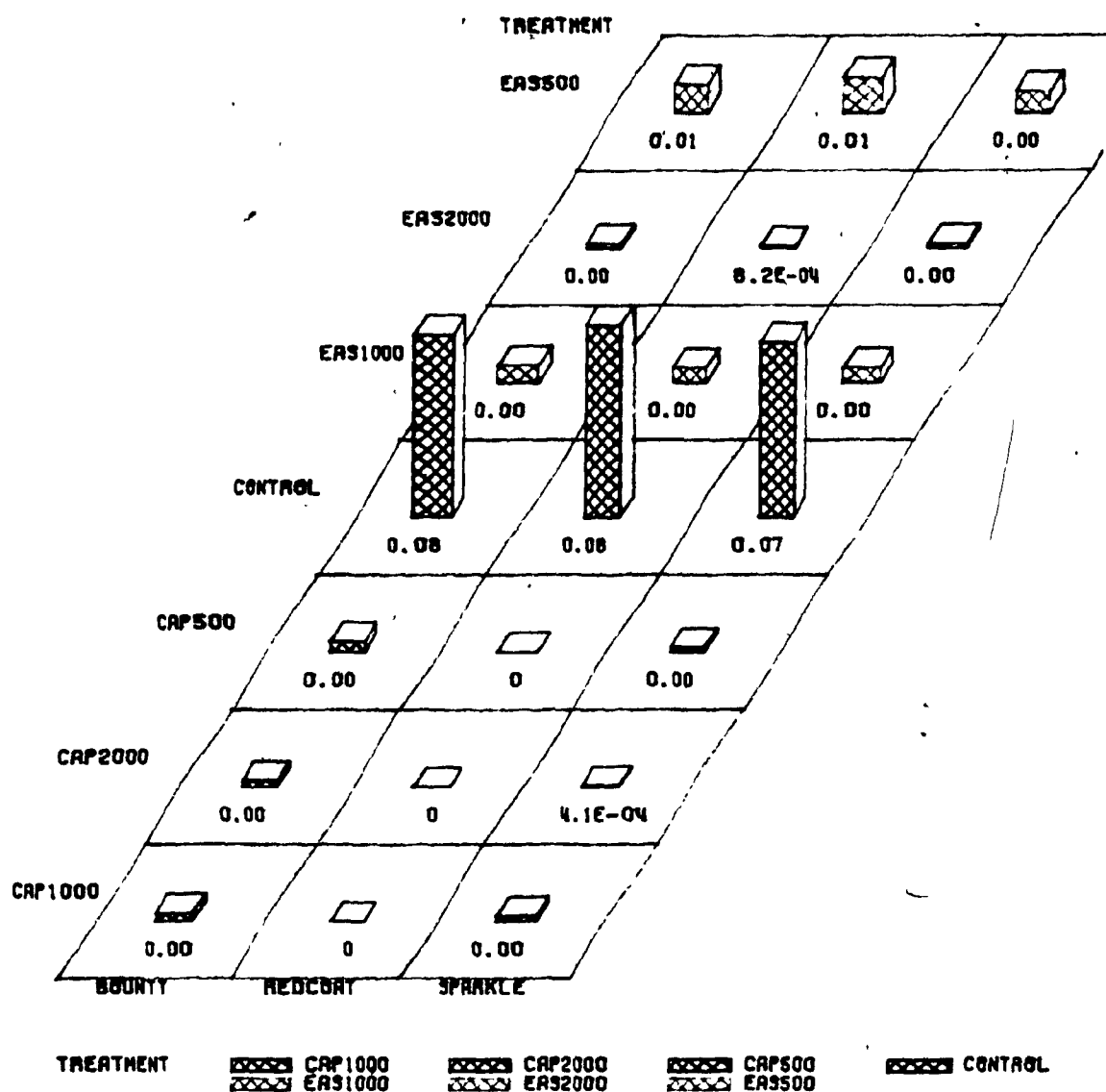


Figure 4. Length of pollen tube (mm) (after three hours) of three strawberry cvs. treated with different concentrations of fungicide in the germination media. Each bar represents the mean of three observations. EAS500 = Easout 500 ppm; EAS100 = Easout 1000 ppm; EAS2000 = Easout 2000 ppm. CAP500 = Captan 500 ppm; CAP1000 = Captan 1000 ppm; CAP2000 = Captan 2000 ppm.

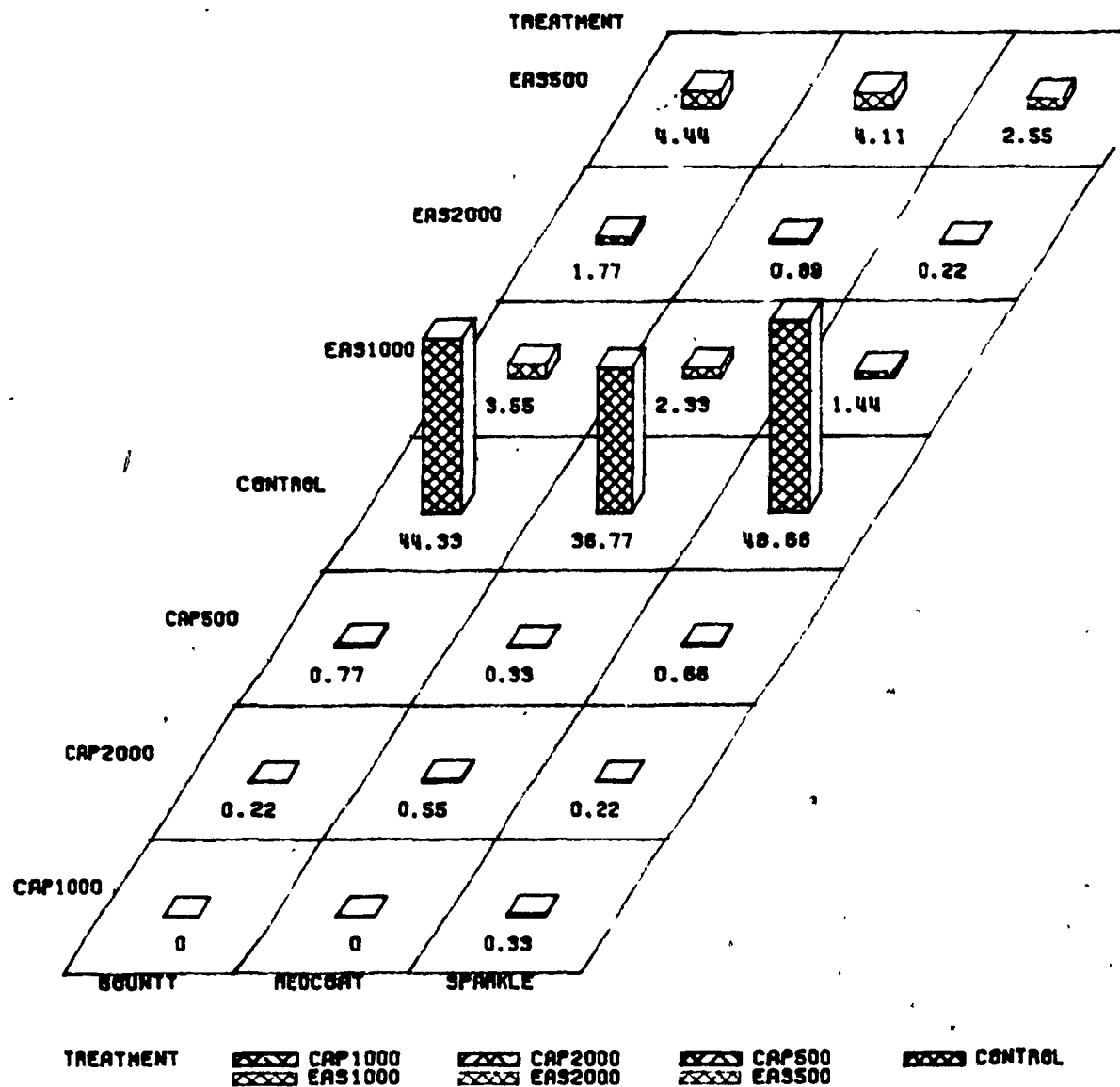


Figure 5. Percent pollen germination (after six hours) of three strawberry cvs. treated with different concentrations of fungicide in the germination media. Each bar represents the mean of three observations. EAS500 = Easout 500 ppm; EAS1000 = Easout 1000 ppm; EAS2000 = Easout 2000 ppm; CAP500 = Captan 500 ppm; CAP1000 = Captan 1000 ppm; CAP2000 = Captan 2000 ppm.

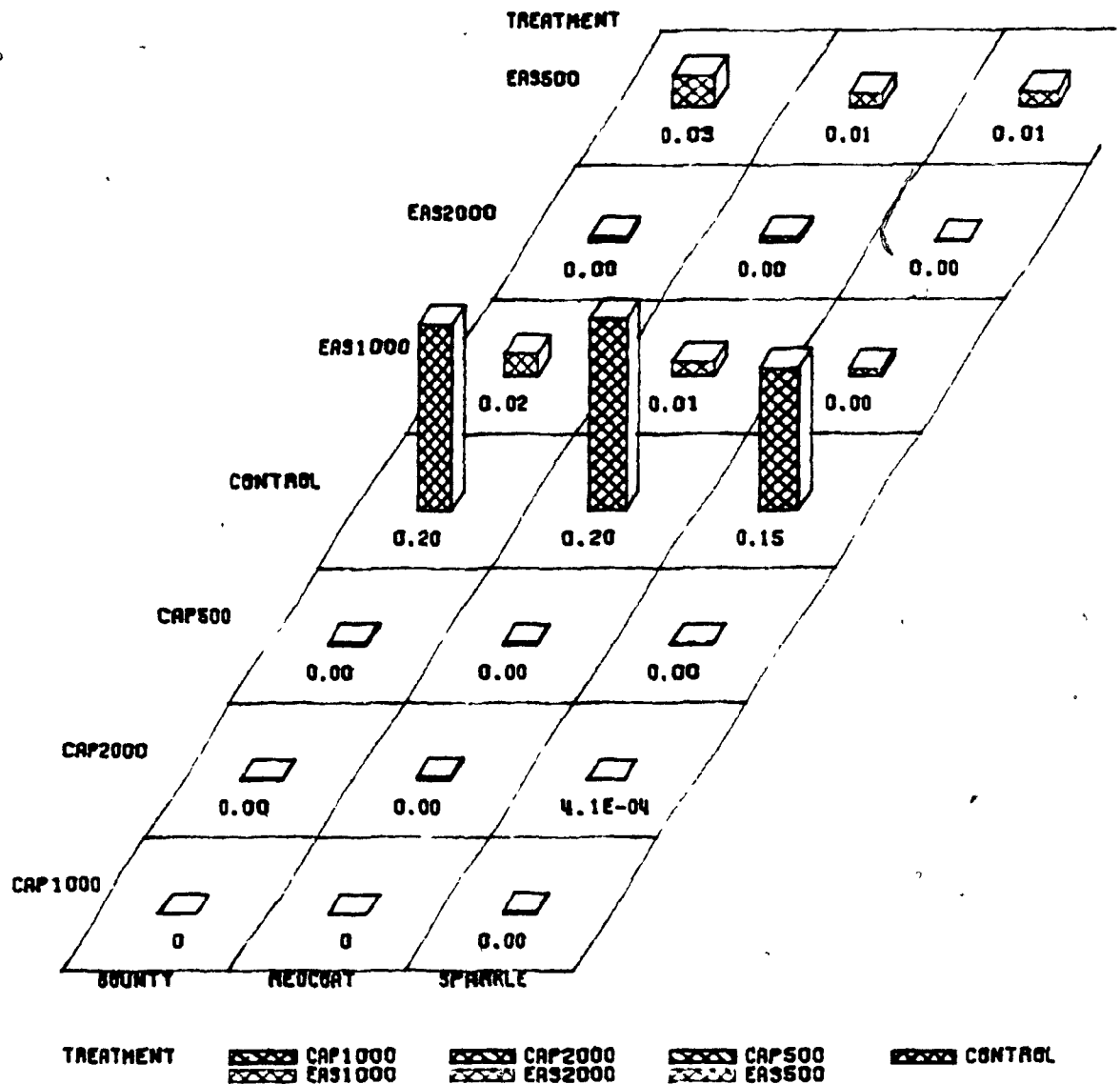


Figure 6. Length of pollen tube (mm) (after six hours) of three strawberry cvs. treated with different concentrations of fungicide in the germination media. Each bar represents the mean of three observations. EAS500 = Easout 500 ppm; EAS1000 = Easout 1000 ppm; EAS2000 = Easout 2000 ppm; CAP500 = Captan 500 ppm; CAP1000 = Captan 1000 ppm; CAP2000 = Captan 2000 ppm.

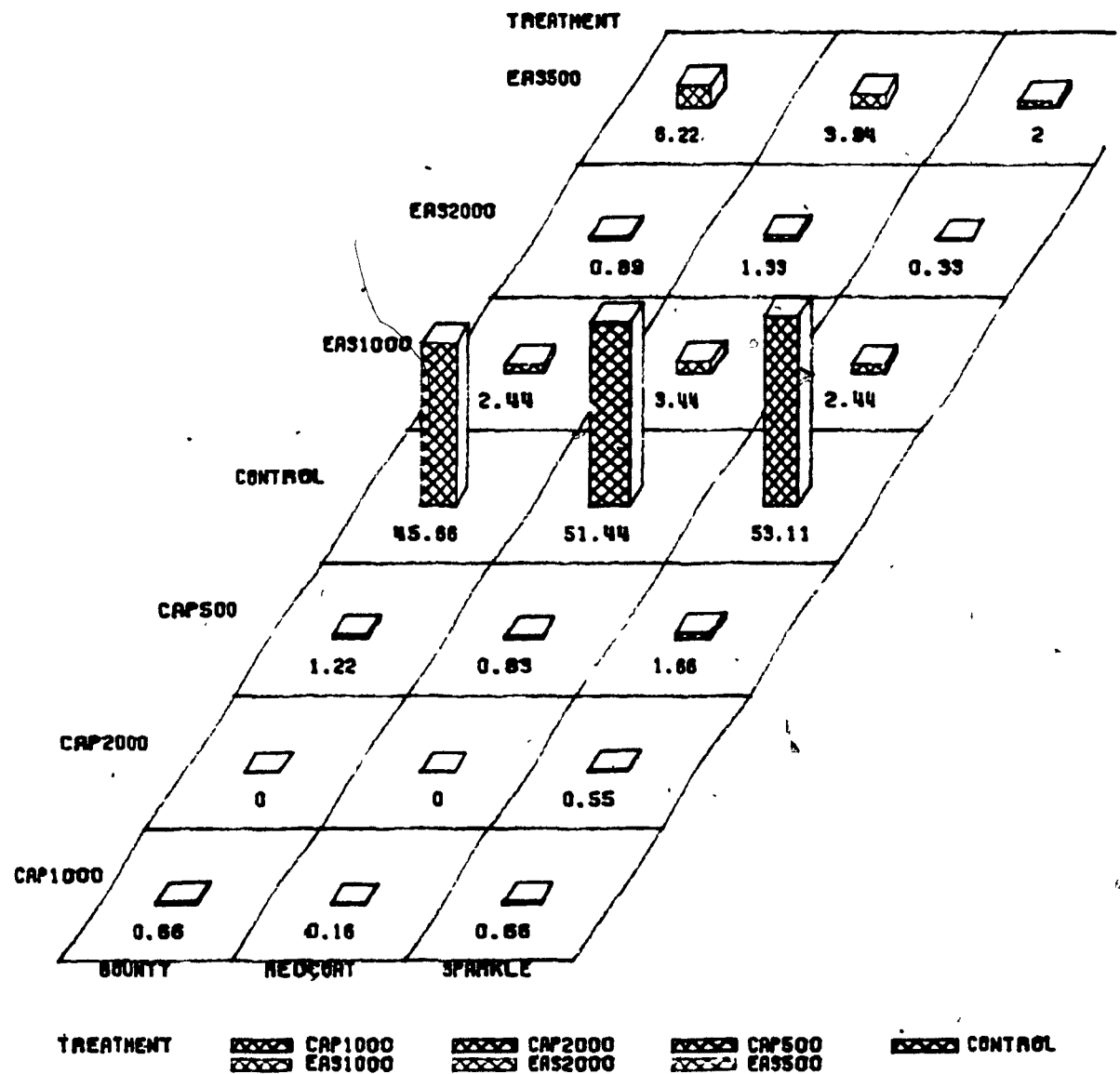


Figure 7. Percent pollen germination (after nine hours) of three strawberry cvs. treated with different concentrations of fungicide in the germination media. Each bar represents the mean of three observations. EAS500 = Easout 500 ppm; EAS1000 = Easout 1000 ppm; EAS2000 = Easout 2000 ppm; CAP500 = Captan 500 ppm; CAP1000 = Captan 1000 ppm; CAP2000 = Captan 2000 ppm.

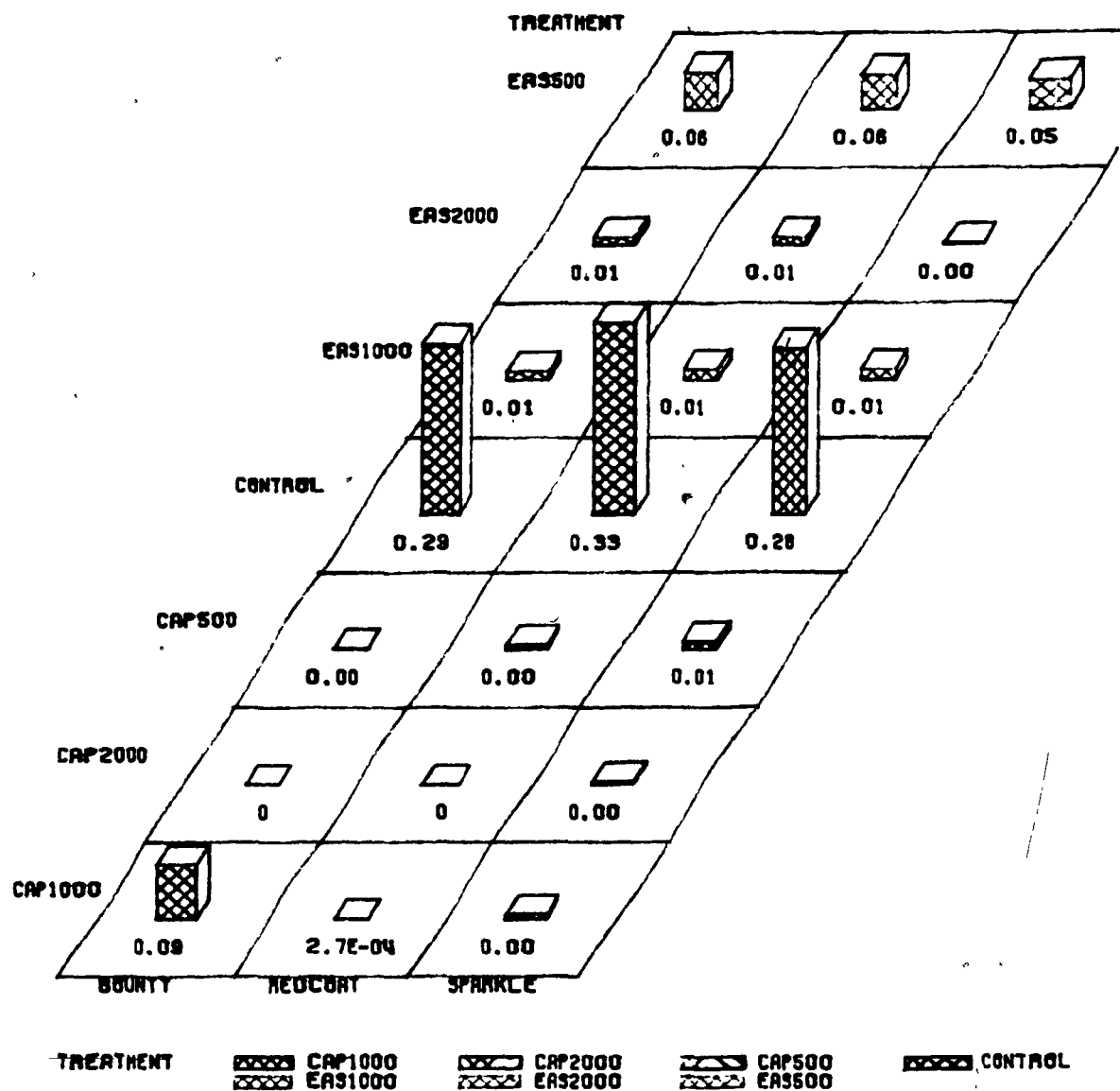


Figure 8. Length of pollen tube (mm) (after nine hours) of three strawberry cvs. treated with different concentrations of fungicide in the germination media. Each bar represents the mean of three observations. EAS500 = Easout 500 ppm; EAS1000 = Easout 1000 ppm; EAS2000 = Easout 2000 ppm; CAP500 = Captan 500 ppm; CAP1000 = Captan 1000 ppm; CAP2000 = Captan 2000 ppm.

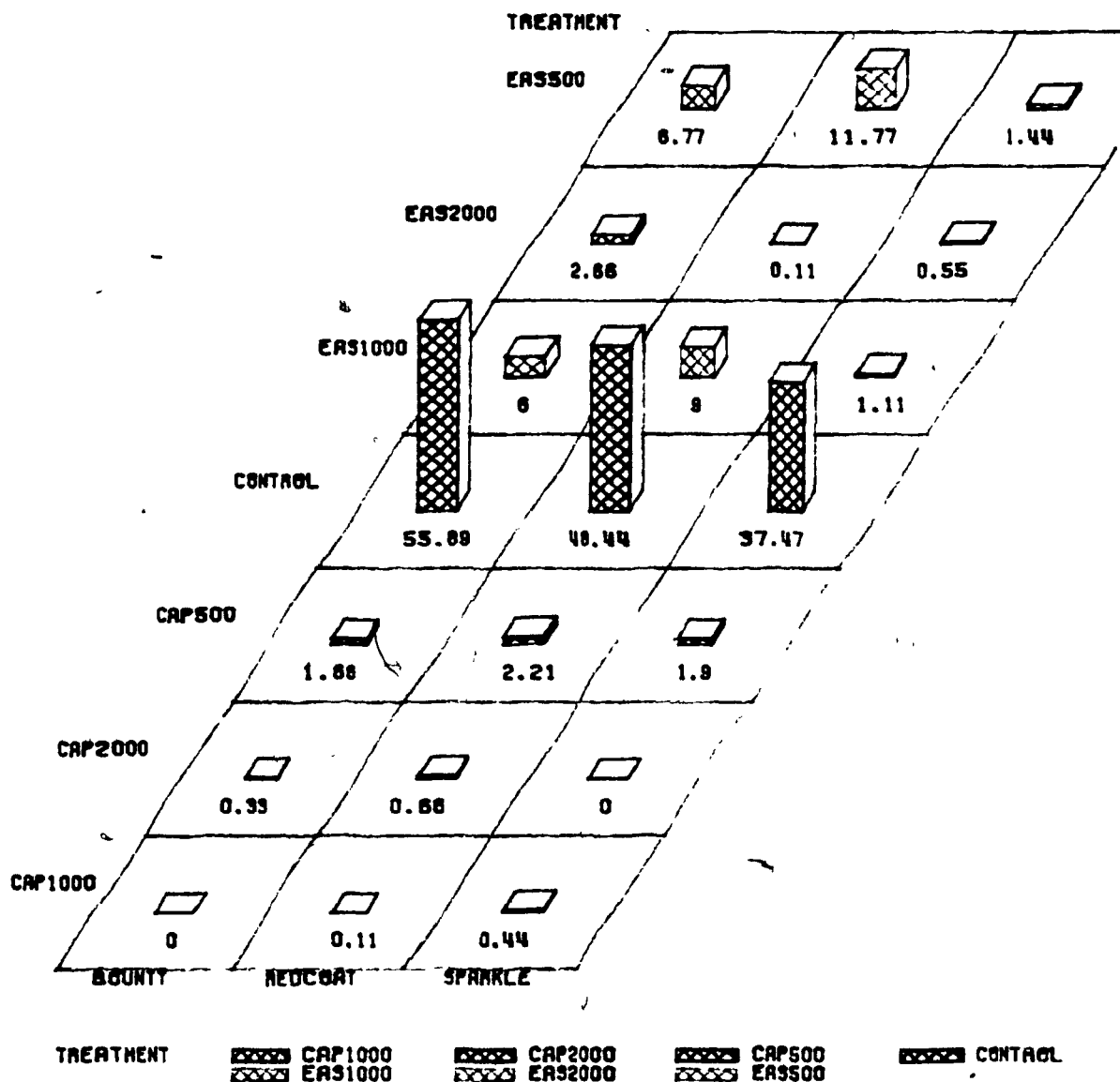


Figure 9. Percent pollen germination (after 24 hours) of three strawberry cvs. treated with different concentrations of fungicide in the germination media. Each bar represents the mean of three observations. EAS500 = Easout 500 ppm; EAS1000 = Easout 1000 ppm; EAS2000 = Easout 2000 ppm; CAP500 = Captan 500 ppm; CAP1000 = Captan 1000 ppm; CAP2000 = Captan 2000 ppm.

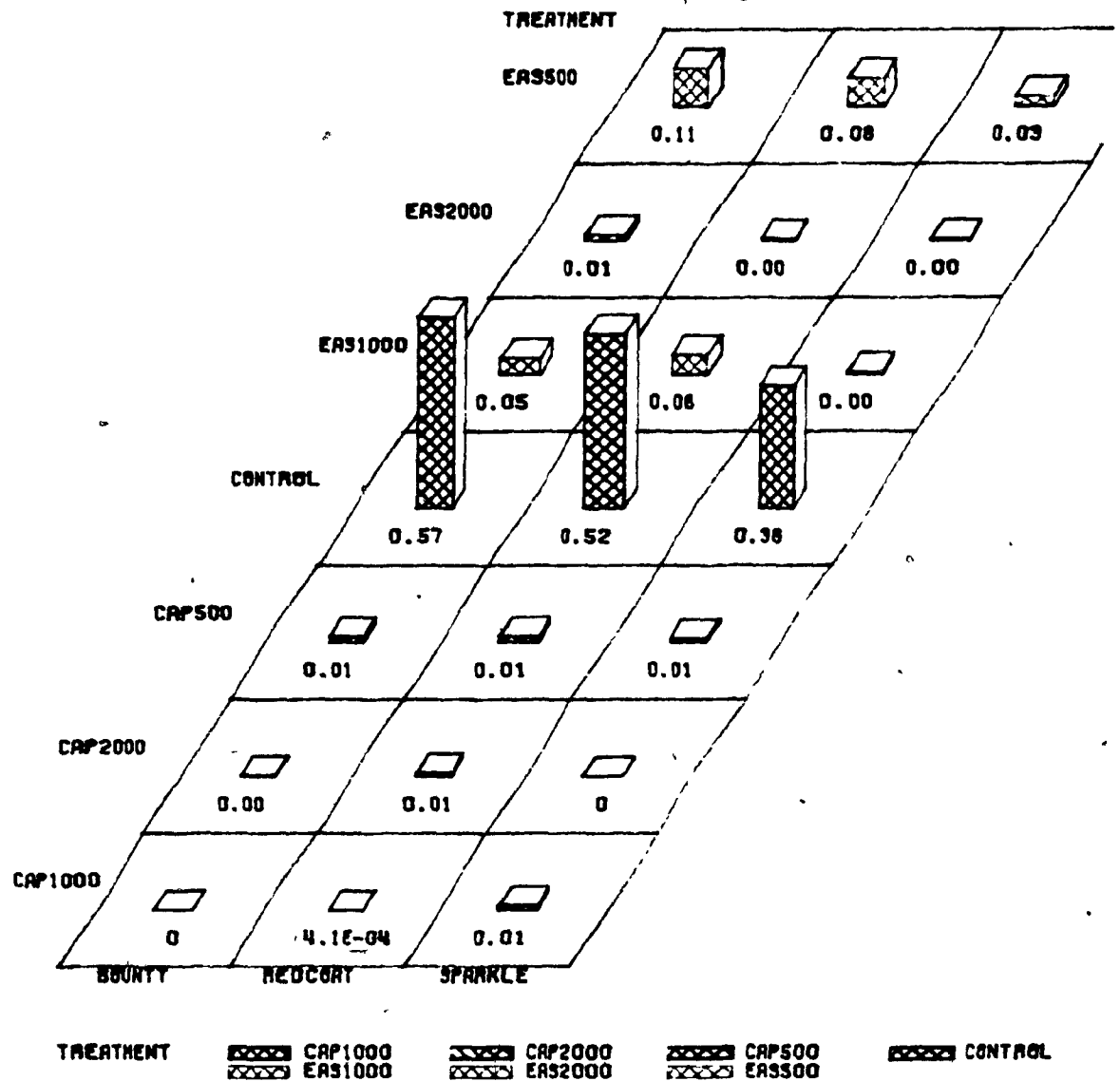


Figure 10. Length of pollen tube (mm) (after 24 hours) of three strawberry cvs. treated with different concentrations of fungicide in the germination media. Each bar represents the mean of three observations. EAS500 = Easout 500 ppm; EAS1000 = Easout 1000 ppm; EAS2000 = Easout 2000 ppm; CAP500 = Captan 500 ppm; CAP1000 = Captan 1000 ppm; CAP2000 = Captan 2000 ppm.

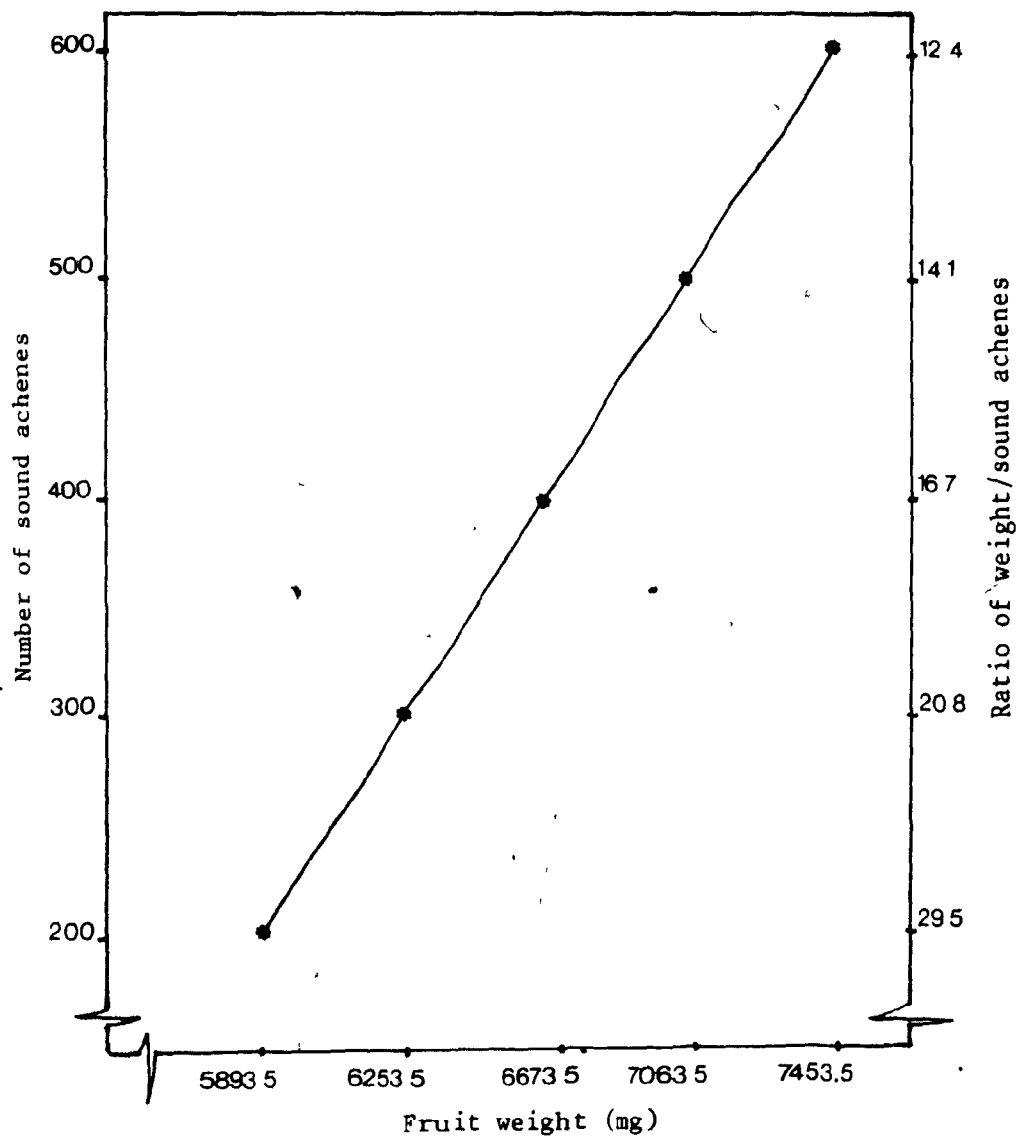


Figure 11. Regression of mean fruit weight (mg) on number of sound achenes.

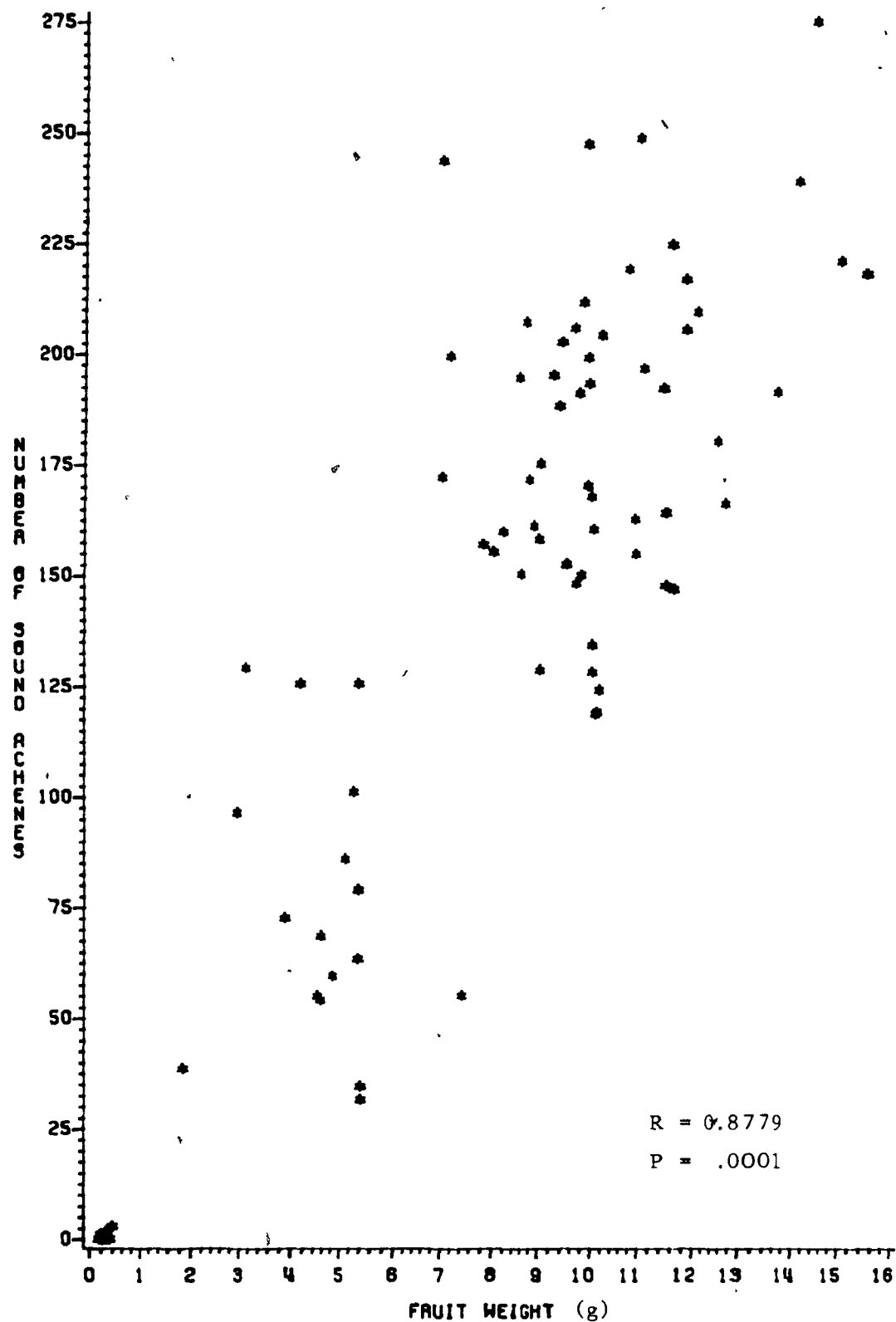


Figure 12. Scatter diagram to illustrate correlation between the number of sound achenes and fruit weight (Experiment 2).