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PREDICTING VECTOR OCCURRENCES AND DISEASE INCIDENCE IN TOMATO CROPS: A CONTROL STRATEGY.

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ABSTRACT

Tomato big bud is caused by a phloem-restricted mycoplasma-like organism. It is an economically important disease of transplant processing tomato crops, causing serious losses in yield in some seasons, in an unpredictable manner. The reported vector of the causal agent of this disease is the common brown leafhopper, Orosius argentatus (Evans) (Homoptera: Cicadellidae), a species widely distributed on a range of weed plants throughout Australia. A review of the disease and the vector in Australia is given. A computer model, based on the biology of the vector, using the method of degree days above the threshold temperature, was developed to predict the various life stages of the insect. These predictions were compared with actual vector occurrences in the crop and related to disease incidence. The results indicate that it is likely that the adults which overwintered as nymphs, move into the crop. If the prediction is correct, then the routine sampling of plants and insects for the disease agent, using a rapid detection method would be desirable. This would allow for the determination of an "infectivity index" for the overwintering nymphal population to predict the likelihood of a severe disease outbreak in the summer. This would then enable the formulation of control strategies for the vector and the disease from one season to the next.

INTRODUCTION

The Disease

a) Historical Background

For many years, leafhopper-borne plant disease agents have been recognised in Australia, although at the time when the diseases were first reported, the specific vector or vectors and the causal agents of the diseases were not always known. The earliest recorded plant disease reported in Australia to be later associated with a leafhopper vector, was rosette of tomato (Grylls 1979), first described by Cobb (1902). Some time later, this disease was reported from Victoria as tomato blue-top (Strickland 1930). Samuel *et al.* (1933) renamed the disease as tomato big bud, when they demonstrated that the causal agent could be transmitted by budding and grafting, and suggested that the disease could be a virus.

The causal agent of big bud disease continued to be thought of as a virus for many years (Hill 1937, Hill and Mandryk 1954, Helms 1957a, 1957b, 1962) and Smith (1957) considered it to be closely related to cranberry false blossom. However, Doi *et al.* (1967) found that mulberry dwarf, a disease with similar etiology is caused by mycoplasma-like organisms, and more recent work by Bowyer *et al.* (1969) provided evidence that the causal agent of tomato big bud is a mycoplasma-like organism (MLO).

Tomato big bud has caused considerable economic losses in commercial processing tomato crops. It is common in inland areas of New South Wales and Victoria, but is seldom a problem in coastal areas (Anon 1972). Victoria grows about 75% of the nation's processing tomatoes, which

amounted to 113,041 tonnes in the 1985-86 season, with an estimated value of some $11.47 million. In the 1979-80 season, there was a severe outbreak of big bud disease in Victoria, and only an estimated 112, 481 tonnes of fruit was harvested, with an estimated value of $9.17 million. In that season some growers lost their entire crop to tomato big bud disease. The average loss of production in Victoria due to the disease in that year was estimated to be about 30% with a cash loss of about $2.30 million.

b) Symptoms

The characteristic symptoms of the disease are abnormalities in the floral parts; the youngest fruit truss becomes upturned instead of recurved; various forms of overgrowth, adhesion and virescence occur; greening and distortion of floral members occur; stems just below the apices become swollen; purple anthocyanins can accumulate along the veins of the calyces, on the undersides of young leaves and on the youngest portion of the stem. The infected plant in its later stages of growth has a tufted appearance due to the shooting of axillary buds, production of dwarfed leaves and the gradual thickening of stems.

Many MLO diseases are characterized by shoot proliferation, but some MLOs do not cause these symptoms. Also, some shoot proliferations can be associated with the feeding of eriophyid mites (May and Webster 1958, Stubbs and Meagher 1965, Shykhui 1969).

c) Plant Diseases with Associated Mycoplasma-like Bodies

There have been many reviews on mycoplasma-like agents in plants (Whitcomb and Davis 1970, Whitcomb 1973, Maramarosch et al. 1970, Whitcomb 1981, Whitcomb and Black 1982, Maramarosch and Raychaundhuri 1979, Hull 1971, Davis and Whitcomb 1971, Hampton 1972, Ghosh and Raychaundhuri 1972, Purcell 1982). When one subtracts from these lists of diseases those now associated with the walled organisms or spiroplasmas, one is left largely with the classical yellows-type diseases of which aster yellows is a prototype (Chykowski 1973).

At present there are some 30 kinds of plant diseases known to be caused by MLOs that are transmitted by leafhoppers (Hull 1971, Maramarosch et al. 1970, Nielson 1968, Iida 1978).

In Australia, tomato big bud, legume "little leaf", lucerne witches' broom, purple top wilt of potato, clover phyllody, strawberry little leaf, and papaw yellow crinkle are all thought to be diseases of the "yellows" type (Bowyer and Atherton 1970, 1971a, Bowyer 1974, Norris 1954, Conroy 1954, Shanmuganathan and Garrett 1976, Stubbs 1968, Greber 1966, Grylls 1979).

Shanmuganathan and Garrett (1976), successfully transferred purple top wilt of potato, clover phyllody and tomato big bud to strawberry using dodder (Cuscuta campestris), resulting in typical symptoms of little leaf in strawberry. They also demonstrated symptoms of big bud in tomato by transferring the pathogens causing clover phyllody and purple top wilt of potato to tomato by dodder.

Other diseases in Australia known to be associated with MLOs are: big bud of tobacco (Hill 1937) and rubbery wood of apple (Washington and Nancarrow 1983, Beakbane et al. 1971).

Some diseases in Australia suspected of being associated with MLO's are citrus die-back (Fraser and Broadbent 1979) and Australian vine yellows (Rhine riesling problem or Chardonnay decline) (Magarey et al. 1983).

Symptoms in plants presumed to be caused by the tomato big bud causal organism have also been reported in various ornamentals, native plants and weeds in Victoria, as well as in various fruits, vegetables, field crops and pasture plant species (Chambers 1982, Washington and Nancarrow 1983, Woodcock and Clarke 1983).

The tomato big bud disease has also been reported from India (Varma 1979), the USA (Dale and Smith 1975, Garnett and Provvidenti 1974) and is known as Stolbur in the USSR (Bagdasaryan and Panteleev 1972, Khar'Kova 1979, Ploaie 1972, Sukhov and Vovk 1951).

The Vector

a) General

The reported vector for tomato big bud in Australia is the common brown leafhopper, Orosius argentatus (Evans), formerly known as Thamnotettix argentata Evans. It is also reported to be the vector for such "yellows" diseases as legume "little leaf", lucerne witches' broom and possibly potato purple top wilt (Hill 1943, Helson 1951, Hutton and Grylls 1956, Norris 1954). This leafhopper is also reported to be the vector of tobacco yellow dwarf (Hill 1940, 1950, Helson 1942, Hill and Mandryk 1954) and bean summer death diseases, which were also thought to be MLO diseases, but are now known to be caused by viruses (Bowyer and Atherton 1971a, Thomas and Bowyer 1979). The vector is widely distributed throughout Australia and the Australian region (Hill and Helson 1949). It is said to occur on many Pacific Islands, the Philippines and Malaysia and was probably introduced into Australia from India (Oman 1949).

b) Life cycle

The life cycle of the leafhopper is described by Helson (1942). Oviposition can commence 3 to 7 days after adult eclosion and occur any time of day when the temperatures are greater than 15.6°C. The incubation period is 7 to 22 days. There are 5 nymphal instars. Females fertilized in the autumn can resume oviposition in the spring without further access to males. Insects overwinter as adults in a cold winter, but in warmer districts such as Northern Victoria, the overwintering stages are both adults and late instar nymphs. Three complete generations a year occur, both in the laboratory and the field. The generations overlap, and breeding is continuous.

c) Leafhopper Movements

Very little is known on the factors which influence leafhopper flights and movement into tomato crops. The movement of the vector into the crop seems to be a seasonal occurrence. The influence of weather and the occurrence of summer host plants, could be factors influencing vector migration into crops.

Hill (1941, 1950) reports that the appearance of the leafhopper in tobacco crops "occurs as a result of migration induced by the drying up of the winter and early spring host plants". This period usually occurs in late November. Helson (1942) suggested that the leafhopper does not move tobacco out of preference for that plant. As soon as alternate weed host plants appear, insects leave the tobacco and move back onto weeds. It seems that if weed hosts do not die or dry up then the insect has little tendency to move away from them. Helson (1950) and Hill and Mandryk (1954) also reported that migration is induced by the drying up of winter-spring host plants. Helson (1951) reported seasonal abundance of leafhoppers with a spring peak for O. argentatus by early November.

d) Leafhopper Feeding

Day et al. (1952) and Day and McKinnon (1951) studied some aspects of the feeding of O. argentatus. Day et al. (1952) reported that the leafhopper was a phloem feeder and that it finds the tissue upon which it feeds by random probing with stylets. The insect does feed on parenchyma cells, but feeds longer on phloem. The leafhopper will invariably reach phloem tissue if it feeds on the undersurface of the leaf. This position seems to be preferred by the vector; Day et al. (1952) further state that the difference in infectivity rates in crops attacked by leafhoppers may be explained by the varying ability of the leafhoppers to locate tissues in each crop in which the causal agent of the disease could multiply.

e) Seasonal Incidence of Disease in the Crop

Some information has been reported on vector activity and seasonal disease incidence (Samuel et al. 1933, Hill 1937, Helson 1951 and Helms 1957b), which indicate that big bud disease is predominantly a mid-summer to late-summer disease. However, no information has been reported on factors influencing the distribution or abundance of the causal organism of tomato big bud in both perennial and annual weed host plants and how this may influence the severity of disease incidence from season to season.

f) Host plants

O. argentatus has a wide range of host plants, many of which are common weeds. The insect has been observed to complete its life cycle on 19 different species from 11 families, and there are some 15 species from 8 families on which adults feed but do not breed. Two such plants include tobacco and tomato (Helson 1942). Hill (1943) further lists 23 plants to which the "big bud" pathogen was experimentally transferred to by the leafhopper. Hutton and Grylls (1956) list 10 species to which "little leaf" disease was transmitted by O. argentatus. They also list 28 species and strains of legumes which are naturally infected with "little leaf" disease as well as 46 species and strains of legumes with field resistance to "little leaf" infection. Hill and Mandryk (1954) report Nicotiana glauca Graham as a symptomless carrier of both "big bud" of tomato and "yellow dwarf" of tobacco diseases. Fragaria vesca L. (strawberry) and Pisum sativum L. cv. 75 (pea) are also reported by Helms (1962) as host plants of "big bud" disease.

Osmelak (1984) gives a list of vector host plants as well as indicating those plants which are also hosts to the pathogens transmitted by O. argentatus. Many of the plants on which the vector can complete its life cycle are also host plants for the "big bud" MLO.

Other Vectors

The specific insect vectors for plant MLO's belong to the superfamilies Cicadelloidea, Fulgoroidea, Cercopoidea and Psylloidea (Osmelak 1984). Day and Bennetts (1954) and Neilsen (1968) detail many leafhopper vectors of phytopathogenic viruses (several of which are now known to be MLOs). However, some of these insects do not occur in Australia.

Very little work has been done in Australia on the vector status of various insects for specific MLO diseases. Grylls (1979) lists various cicadellid and delphacid vectors of plant disease agents in Australia, but these are restricted to very few species for MLO diseases. O. argentatus has either been proven to be a vector for some MLO diseases on the basis of laboratory transmission experiments, or implicated to be a vector of some MLO diseases by association with other 'yellows' diseases. This, however, leaves some doubt as to whether this leafhopper is in fact the main vector of some diseases in years of severe disease incidence in the field.

TABLE 1.

Genera of Cicadelloidea and Fulgoroidea known to transmit plant pathogens, and which also occur in Australia.

	Genus	Source
CICADELLOIDEA		
Cicadellinae	Cicadella	Kent (1955)
Deltocephalinae	Cicadulina	Nagpal et al. (1978)
	Deltocephalus*	Ploaie et al. (1977)
	Empoasca*	Sein and Adsuar (1947)
	Euscelis*	Stanarious et al. (1977)
	Hishimonous	Sakai (1937)
	Nephotettix*	Anon (1977)
	Nesoclutha	Greber (1977)
	Orosius*	Nagaich et al. (1974)
	Recilia*	Anon (1977)
	Scaphoideus*	Belli & Osler (1977)
Macropsinae	Macropsis	Oberle & Wingard (1967)
Agallinae	Austroagallia	Grylls et al. (1974)
FULGOROIDEA		
Delphacidae	Nilaparvata	Abeygunawardena et al.(1971)
	Peregrinus	Greber (1977)
	Perkinsiella	Gollifer et al. (1977)
	Sogata	Trujillo (1969)
	Sogatella	Mariappan et al. (1975)
	Sogatodes	Lobaton & Martinez (1977)
	Tarophagus	Gollifer et al. (1977)
	Toya	Harder & Bakker (1974)
Cixiidae	Oliarus*	Ushiyama et al. (1969)

* Vectors of MLO diseases.

It would therefore seem advisable to keep in mind the insects which are known to transmit plant pathogens as any one of them may prove to be an alternate vector of the tomato big bud agent. The genera of the Cicadelloidea and Fulgoroidea superfamilies, which are known to occur in Australia (Table 1) have been extracted from reports of known vectors of plant pathogens around the world. The list is not exhaustive but does help illustrate the potential for vectors of MLO diseases in Australia other than O. argentatus.

Other vectors of tomato big bud or stolbur are given by Ishihara (1969), Neilson (1968) and Bagdasarayan and Panteleev (1972). Ishihara (1969) however does not give his source of information, and most of the vectors listed are mentioned by Neilson (1968). Many of the vectors listed do not occur in Australia (M. Fletcher, pers. comm.) Moreover, three of the vectors listed by Ishihara (1969) namely Empoasca devastans Dist., Hishimonous phycitis Dist. and Peragallia sinuata (Muls and Rey), have not been shown conclusively to be vectors of the disease (Neilson 1968).

Control

Control methods for plant MLO diseases can be directed against the disease agents or against the specific vectors (Maramarosch 1963). Many methods have already been tried including breeding for resistance, antimycoplasmal drugs, heat therapy, plant surgery, screens to eliminate vectors and control of vectors by biological and chemical means.

At present, the only form of control attempted for tomato big bud in Australia has been chemical control of the vector. The application of foliar sprays for the control of leafhoppers in tomato crops has not been very successful. However, some systemic insecticides have been reported to reduce the incidence of leafhopper borne diseases (Georghiou *et al.* 1964, Malm and Finker 1968, Paddick and French 1964, 1972, Peay and Oliver 1964, Thomson and Rawlins 1961, Howard and McKoy (1980).

Tetracycline antibiotics are known to suppress development of symptoms of diseases associated with MLOs (Davis *et al.* 1968, Ishiie *et al.* 1967, Bowyer and Atherton 1972). However the use of antibiotics in tomato crops would not be economical.

The Research Program

As very little information existed on the ecology of *O. argentatus*, a monitoring program was designed to investigate various aspects of leafhopper activity and occurrence, especially from the time of seeding to harvest of the tomato crops. This information was then related to disease incidence in tomato crops. The objectives of the program were to further the understanding of the epidemiology of the disease, as well as to develop a prediction model for vector movements into the crop. This would help in formulating control measures for the leafhopper vector and tomato big bud disease.

This paper reports only on aspects of leafhopper monitoring in the crop and the leafhopper phenology model.

METHODS

The experimental plots consisted of 3 rows 15.24 metres in length and 1.52 metres apart, with a buffer row between plots in six bays at each trial site.

During the 1980/81, 1981/82 and 1982/83 seasons, leafhoppers were trapped in the crop, using the sweep net method. 50 sweeps per plot were taken once a week, with a 38 cm diameter net.

Pan traps filled with ethylene glycol were used as well as sweep nets during the 1982/83 season. Only pan traps were used during the 1983/84, 1984/85, 1985/86 seasons. Sweep net samples and pan trap contents were collected every 7 days.

To determine disease incidence, the appearance of disease symptoms as described by Samuel *et al.* (1933) was monitored weekly by inspecting each plant in each plot.

A computer model, based on the known biology of *O. argentatus* as reported by Helson (1942), was developed to predict the various life stages of the insect at any point in time. The phenology model performs thermal integrations on daily maximum and minimum temperature data, to simulate the life stages of the leafhopper vector. The thermal integrations follow those of Allen (1976), but were modified to allow for a more sophisticated use of upper and lower thresholds.

The leafhopper life cycle is divided into stages (Table 2). For each stage, the day degrees required, the upper and lower thresholds, the name and whether a vertical or horizontal cut off is to be used, and the base rate of development for any stage may be entered.

Table 2. Leafhopper Life Stages & Temperature Requirements

STAGE	STAGE NAME	DDREQ.	T.BASE	T.HIGH	T.LIMIT	V.CUTOFF
1	Egg (Adult in trap)	113.00	13.70	32.00	13.70	F
2	First Instar	51.00	12.80	32.00	12.80	F
3	Second Instar	51.00	12.80	32.00	12.80	F
4	Third Instar	51.00	12.80	32.00	12.80	F
5	Fourth Instar	51.00	12.80	32.00	12.80	F
6	Fifth Instar	51.00	12.80	32.00	12.80	F
7	Pre-Mating Adult	40.00	15.60	32.00	15.60	F

DDREQ. Day-degrees required.
T.BASE Lower development threshold ^{o}C.
T.HIGH Upper development threshold ^{o}C.
T.LIMIT Temperature (^{o}C) at which no development occurs. In this case = T.BASE
V.CUTOFF Vertical Cutoff. Temperature ^{o}C where development ceases due to death
F. No vertical cutoff (so far THE OPTION for V.CUTOFF has not been used).

The simulation is driven by maximum and minimum temperature data. Simulations may go forward or backward in time. If a forwards simulation extends beyond the available temperature data, the temperature is calculated on 12 corresponding periods over 1971 to 1982. The events described in that case, represent the mean time and standard deviation of the event, based on the 12 simulations. Backward simulations are performed on the mean data for the projected period.

In order to start the model one assumption was made. This was that 5 consecutive days above the pre-mating threshold of 15.6^{o}C had to occur before overwintering adults recommenced oviposition. On that basis, the model was selected to start on September 9, 1980, and simulations were projected till February 1986. The predictions of adult emergences were then compared to leafhopper movements into transplant tomato crops.

Some validation of the model was attempted by placing pitfall traps at various trial sites to detect the different life stages of the leafhopper vector. Trap contents were collected every 7 days.

RESULTS

The results for the number of O. argentatus caught in the crop at each trial site for the seasons from 1980 to 1986 are illustrated in Figs 1 & 2. The results indicate that the vector moves into the crop early in the season. The pan trap method caught more leafhoppers than the sweep net method, but both methods reflect the same trend in leafhopper occurrences in the crop.

The incidence of disease during the monitoring period never exceeded 5%. No relationship between the total number of leafhoppers found in the crop, and disease incidence in the crop could be determined from the data. The incidence of disease peaked some 8 weeks or more after the influx of leafhoppers into the crop.

A comparison of the predicted adult leafhopper stages with the time when leafhoppers move into transplant crops is also illustrated in Figs. 1&2. This shows that the majority of adults found in the crop, could be those which actually overwintered as nymphs.

Only a few results were obtained from the pitfall traps. Apart from adult leafhoppers, only 5th instar nymphs were detected on two occasions. Each time, the immature leafhoppers were trapped three days prior to the predicted emergence of 5th instar nymphs.

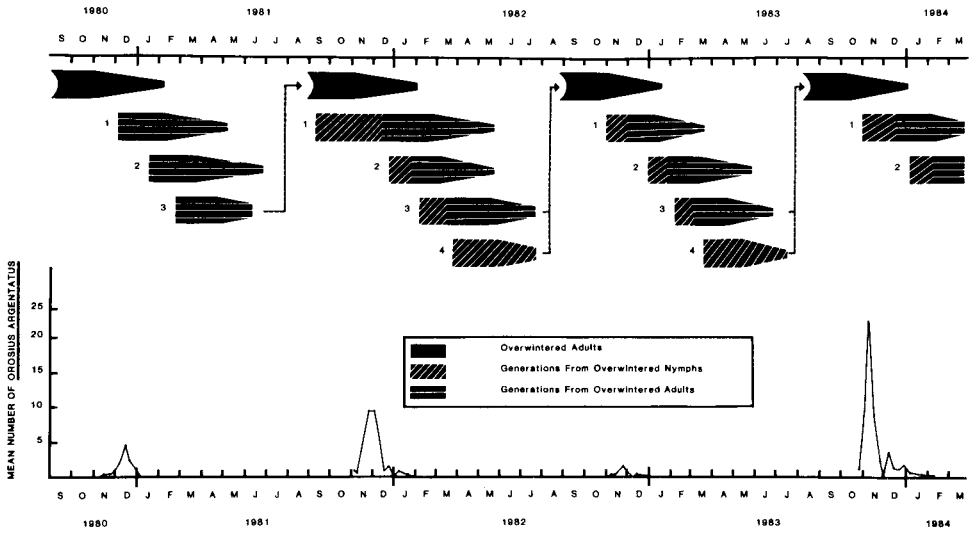

Fig 1. Comparison of forecasted adult stage of Orosius argentatus and occurrence of adults in tomato crops. 1980-1984.

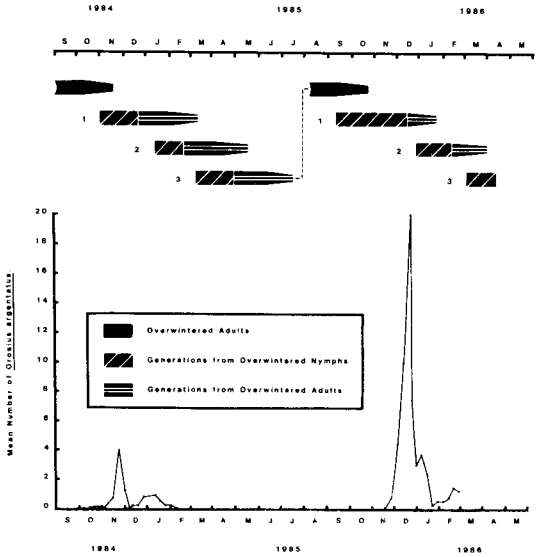

Fig 2. Comparison of forecasted adult stage of Orosius argentatus and occurrence of adults in tomato crops. 1984-1986.

DISCUSSION

Disease incidence in the crop peaked some 8 weeks or more after the influx of leafhoppers in the crop. Symptoms of the disease cannot be confirmed in the field less than 6-8 weeks after innoculation. These results suggest therefore, that disease transmission occurred in the crop and not in the seedbed for the 1980-84 seasons.

The results using the computer model, have been encouraging enough to justify more effort being devoted to refining the model. If it is the adults which overwintered as nymphs that are responsible for the disease in the crop, it may be possible to predict when leafhoppers move into the crop. However, it will still not be possible from that to predict the percentage disease incidence that is likely to occur.

If the phenology prediction is correct, then a rapid detection method for the routine sampling of plants and insects for the disease causal agent, would be desirable. This would allow for the determination of an "infectivity index" for the overwintering nymphal population and could serve as a guide in predicting the likelihood of a severe disease outbreak in the summer. Such a method is as yet not available for the causal agent of tomato big bud. The procedures described by Chapman (1971,1985) to obtain an "infectivity index" for the control of the aster yellows problem, is considered to be too time-consuming for tomato big bud. Also, the tomato big bud problem in Australia does not have the benefit of 4 decades or more of continuous research, as for the aster yellows problem, which can be drawn upon for a greater understanding of the epidemiology of the disease.

Enzyme-linked immunosorbent assay (ELISA) methods for a few plant mycoplasma diseases of the "yellows" type have been developed (Clarke 1981) and preliminary work to develop an ELISA test for the tomato big bud mycoplasma has already begun. The development of a rapid detection method is probably the most important area for future research. If such a test can be developed, then a sampling of weed host plants on which the overwintering nymphal population are feeding, as well as other weed plants known to be host to the disease causal agent and the general leafhopper population, could be made. This would lead to the calculation of an "infectivity index" which could then be used as a guide to disease incidence in the summer of the coming season. A prediction service to notify growers when leafhoppers are likely to move into the crop and what incidence of disease is expected, would then be possible.

The majority of the known MLO diseases in Australia have also been shown by dodder transmission tests to give the same symptoms of tomato big bud in tomatoes. All of these diseases are thought to be transmitted by the same leafhopper which is the vector of tomato big bud disease. This phenology model may therefore also serve to help formulate control methods for these other MLO problems.

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