

SOME VECTOR, VIRUS, HOST-PLANT RELATIONSHIPS OF THE  
SIX-SPOTTED LEAFHOPPER, Macrosteles fascifrons (Stal)<sup>o</sup>  
AND ASTER YELLOWS VIRUS IN MANITOBA.

A Thesis

Presented to

The Department of Entomology  
Faculty of Agriculture and Home Economics  
The University of Manitoba

In Partial Fulfillment  
of the Requirements for the Degree  
Doctor of Philosophy

by

Howard Percival Richardson

January 1966



ABSTRACT

by

Howard Percival Richardson

SOME VECTOR, VIRUS, HOST-PLANT RELATIONSHIPS OF THE  
SIX-SPOTTED LEAFHOPPER, Macrosteles fascifrons (Stål<sup>o</sup>),  
AND ASTER YELLOWS VIRUS IN MANITOBA.

Contact and systemic insecticides formulated as emulsive concentrates (EC), wettable powders (WP), and granules (G) were compared at various times, rates and intervals of application to control the six-spotted leafhopper, Macrosteles fascifrons (Stål<sup>o</sup>), and prevent the spread of aster yellows virus (AYV) to head lettuce and carrots. The insecticides malathion EC, Baygon EC and G and phorate G, at the rate of one pound per acre controlled the six-spotted leafhopper and prevented the spread of AYV to the head lettuce in the spring and summer crops of 1960, 1961 and 1962. The same insecticides gave only partial protection to the spring crop and failed to protect the summer crop of 1963 because of a combination of a large population of M. fascifrons with a high percentage (ten per cent) of infective leafhoppers. Other insecticides tested against M. fascifrons on head lettuce were less effective.

The start of applications of insecticides to the spring crops may be delayed for two or three weeks after the plants emerge. On summer crops, spray applications must begin at crop emergence.

DDT EC and Carbaryl WP, one pound per acre, one application per week gave a significant reduction in carrots affected by AYV when the spray applications were started within two to three weeks of crop emergence.

Less than 0.5 p.p.m. of malathion was found on lettuce heads which were analyzed nine days after the last of 15 applications.

Three strains of AYV, "A", "B", and "C", were isolated from lettuce, zinnia and celery, respectively, in Manitoba. The three strains were separated by symptoms on aster, Nicotiana rustica var. humilis and celery. The criteria: 1) plant height from first leaf node to tip; 2) plant height from first to last exposed leaf node; 3) number of exposed leaf nodes and 4) axillary growth on aster were also successfully used to separate the strains.

The transmission of the three strains of AYV by single infective M. fascifrons showed that the six-spotted leafhopper, male or female, is a consistent and reliable transmitter after a two-day inoculation feed; that aster is a poor indicator of the proportion of infective leafhoppers; that stinkweed and head lettuce are superior indicator plants; and that different species of host plants vary in their susceptibility to different strains.

The acquisition of the three strains of AYV by M. fascifrons from 14 different host plants showed that it depended on the strain of AYV and the host plant.

#### ACKNOWLEDGMENTS

I wish to thank Dr. A. E. Hannah, Director, Research Station, Canada Department of Agriculture, Winnipeg, Manitoba for permission to use the project as a thesis, and Dr. A. G. Robinson, Department of Entomology, University of Manitoba, for accepting the project as a thesis.

Thanks are due also to Mr. P. H. Westdal for doing the final statistical analysis of data presented in Chapters IV, V, VI, and VII; to Dr. L. B. Smith and Dr. W. Ives for advice on statistical methods; to Mr. R. Cheale for photographing the plants in Fig. 8, 10, and 19; to Dr. P. Barker, Dr. W. C. McDonald and others for reviewing the manuscript and Mr. J. Ilchyna, Mr. N. Brandt, Mr. D. Tiltman and Mr. G. Cox for assistance in carrying out the field experiments.

TABLE OF CONTENTS

CHAPTER	PAGE
I. INTRODUCTION. . . . .	1
The problem. . . . .	2
Importance of the problem. . . . .	2
Organization of the thesis . . . . .	3
II. REVIEW OF THE LITERATURE. . . . .	4
Control of insect vectors of plant viruses . . .	4
Vector, virus, host-plant relationships. . . . .	8
III. GENERAL MATERIALS AND METHODS . . . . .	17
IV. EXPERIMENTS ON CONTROL OF <u>M. fascifrons</u>	
DURING 1960 . . . . .	32
Materials and methods. . . . .	32
Results and discussion . . . . .	34
V. EXPERIMENTS ON CONTROL OF <u>M. fascifrons</u>	
DURING 1961 . . . . .	41
Materials and methods. . . . .	41
Results and discussion . . . . .	42
VI. EXPERIMENTS ON CONTROL OF <u>M. fascifrons</u>	
DURING 1962 . . . . .	51
Materials and methods. . . . .	51
Results and discussion . . . . .	52
VII. EXPERIMENTS ON CONTROL OF <u>M. fascifrons</u>	
DURING 1963 . . . . .	56
Materials and methods. . . . .	56

CHAPTER	PAGE
Results and discussion . . . . .	57
VIII. DIFFERENTIATION OF THREE STRAINS OF ASTER YELLOW S VIRUS . . . . .	68
Materials and methods. . . . .	68
Results and discussion . . . . .	68
IX. TRANSMISSION BY <u>M. fascifrons</u> OF THREE STRAINS OF ASTER YELLOW S VIRUS TO FOURTEEN DIFFERENT HOST PLANTS. . . . .	89
Materials and methods. . . . .	89
Results and discussion . . . . .	89
X. ACQUISITION BY <u>M. fascifrons</u> OF THREE STRAINS OF ASTER YELLOW S VIRUS FROM FOURTEEN DIFFERENT HOST PLANTS. . . . .	93
Materials and methods. . . . .	93
Results and discussion . . . . .	93
XI. SUMMARY . . . . .	98
BIBLIOGRAPHY. . . . .	103
APPENDIX A. . . . .	108
Richardson, H. P. and P. H. Westdal. 1963. Control of the six-spotted leafhopper, <u>Macrosteles fascifrons</u> (Stål) and aster yellows on head lettuce in Manitoba. Can. J. Plant Sci. 43:12-17. . . . .	109

APPENDIX A (Cont'd)	PAGE
Richardson, H. P. and P. H. Westdal. 1964. Experiments on control of the six-spotted leafhopper, <u>Macrosteles fascifrons</u> , and aster yellows on head lettuce with contact and systemic insecticides in Manitoba. Can. J. Plant Sci. 44:393-396.. . . . .	115
APPENDIX B . . . . .	119
See list of tables for Appendix B . . . . .	xi

LIST OF TABLES

TABLE	PAGE
<p>I. Per cent of head lettuce plants infected with aster yellows after application of malathion at two lb. per acre at two- and four-day intervals with treatment one beginning at crop emergence against <u>Macrosteles fascifrons</u>, Spring crop 1960. . . . .</p>	37
<p>II. Total and seasonal mean number of <u>Macrosteles fascifrons</u> for three replicates after application of malathion at two lb. per acre at four-day intervals, with treatment one beginning at crop emergence, Spring crop 1960. . . . .</p>	38
<p>III. Percentage of head lettuce plants infected with aster yellows after application of malathion at two lb. per acre at two- and four-day intervals with treatment one beginning at crop emergence against <u>Macrosteles fascifrons</u>, Summer crop 1960. . . . .</p>	39
<p>IV. Percentage of carrot plants infected with aster yellows after application of DDT and Carbaryl, each at two lb. per acre, at six-day intervals with treatment one beginning at crop emergence against <u>Macrosteles fascifrons</u>. . . . .</p>	40
<p>V. Percentage of head lettuce plants infected with aster yellows after application of malathion,</p>	



TABLE	PAGE
Carbaryl and Baygon, with treatment one beginning at crop emergence against <u>Macrosteles fascifrons</u> , Spring crop 1961 . . . . .	46
VI. Total and seasonal mean number of <u>Macrosteles fascifrons</u> after application of malathion, Carbaryl, and Baygon, with treatment one beginning at crop emergence, Spring crop 1961. . . . .	47
VII. Percentage of head lettuce plants infected with aster yellows after application of malathion, Carbaryl and Baygon, with treatment one beginning at crop emergence against <u>Macrosteles fascifrons</u> , Summer crop 1961 . . . . .	48
VIII. Total and seasonal mean number of <u>Macrosteles fascifrons</u> after application of malathion, Carbaryl and Baygon, with treatment one beginning at crop emergence, Summer crop 1961. . . . .	49
IX. Seasonal and mean number of leafhoppers after application of malathion and Carbaryl against <u>Macrosteles fascifrons</u> , Summer crop 1961 . . . . .	50
X. Total and seasonal mean number of <u>Macrosteles fascifrons</u> for each insecticide, Spring crop 1962 . . . . .	53
XI. Percentage of head lettuce plants infected with aster yellows virus, Spring crop 1962. . . . .	54

TABLE	PAGE	
XII.	Total and mean number of <u>Macrosteles fascifrons</u> for each insecticide tested, Summer crop 1962 . . . . .	55
XIII.	Percentage of head lettuce plants examined and found infected with aster yellows virus, Portage la Prairie, Spring crop 1963. . . . .	59
XIV.	Total and seasonal mean number of <u>Macrosteles</u> <u>fascifrons</u> , Portage la Prairie, Spring crop 1963. . . . .	60
XV.	Percentage and number of head lettuce plants examined and found infected with aster yellows virus, Winnipeg, Spring crop 1963 . . . . .	61
XVI.	Total and seasonal mean number of <u>Macrosteles</u> <u>fascifrons</u> per 50 sweeps per treatment, Winnipeg, Spring crop 1963. . . . .	62
XVII.	Percentage and number of head lettuce plants examined and found infected with aster yellows virus, Portage la Prairie, Summer crop 1963 . . . . .	63
XVIII.	Total and seasonal mean number of <u>Macrosteles</u> <u>fascifrons</u> per 50 sweeps per treatment, Portage la Prairie, Summer crop 1963. . . . .	64
XIX.	Total number of <u>Macrosteles fascifrons</u> , adults and nymphs on AC 47031, and check, Portage la Prairie, Spring crop 1963 . . . . .	65
XX.	Mean and LSD for 40 aster plants for each of	

TABLE	PAGE
<p>three criteria and the per cent of plants showing axillary growth, a fourth criterion used to differentiate AYV strains "A", "B", and "C" and check. . . . .</p>	70
<p>XXI. Mean and LSD for 50 aster plants for each of three criteria and the per cent of plants showing axillary growth, a fourth criterion used to differentiate AYV strains "A", "B", and "C" and check. . . . .</p>	71
<p>XXII. Mean and LSD for 25 aster plants for each of three criteria and the per cent of plants showing axillary growth, a fourth criterion used to differentiate AYV strains "A", "B", "C" and check. . . . .</p>	72
<p>XXIII. Total and per cent transmission of three strains of aster yellows virus by infected single <u>Macrosteles fascifrons</u> to fourteen different host plants. . . . .</p>	90
<p>XXIV. Total and per cent transmission of three strains of aster yellows virus to aster by single <u>Macrosteles fascifrons</u> based on acquisition from fourteen different host plants. . . . .</p>	96
<p>XXV. Differential transmission of aster yellows virus strains "A", "B", and "C" based on acquisition</p>	

TABLE

PAGE

by Macrosteles fascifrons from fourteen  
different host plants . . . . . 97

LIST OF TABLES

APPENDIX B

TABLE	PAGE
I. Number of head lettuce plants examined and found infected with aster yellows virus after application of malathion at two pounds per acre, Spring crop 1960. . . . .	120
II. Total number of <u>Macrosteles fascifrons</u> per replicate for seven dates of sweeping on head lettuce, Spring crop 1960. . . . .	121
III. Number of head lettuce plants examined and found infected with aster yellows virus, Summer crop 1960. . . . .	122
IV. Malathion residue on head lettuce nine days after applications ceased . . . . .	123
V. Number of carrot plants examined and found infected with aster yellows virus . . . . .	124
VI. Total number of <u>Macrosteles fascifrons</u> per 50 sweeps per subplot made on seven different dates, Spring crop 1961 . . . . .	125
VII. Number of head lettuce plants examined and found infected with aster yellows virus, Spring crop 1961. . . . .	126
VIII. Total number of <u>Macrosteles fascifrons</u> per	

TABLE	PAGE
	50 sweeps per subplot made on five different dates, Summer crop 1961. . . . . 127
IX.	Number of head lettuce plants examined and found infected with aster yellows virus, Summer crop 1961 . . . . . 128
X.	Total number of <u>Macrosteles fascifrons</u> , fifty sweeps per replicate for four replicates for each date of sweeping, Spring crop 1962. . . . . 129
XI.	Number of heads of lettuce examined and found infected with aster yellows virus per replicate per treatment, Spring crop 1962. . . . . 130
XII.	Total number of <u>Macrosteles fascifrons</u> per fifty sweeps per replicate for one date of sweeping, September 17, Summer crop 1962 . . . . . 131
XIII.	Number of <u>Macrosteles fascifrons</u> per 50 sweeps per plot, Portage la Prairie, Spring crop 1963 . . . . . 132
XIV.	Number of head lettuce plants examined and found infected with aster yellows virus, Portage la Prairie, Spring crop 1963 . . . . . 133
XV.	Number of <u>Macrosteles fascifrons</u> per 50 sweeps per plot, Winnipeg, Spring crop 1963 . . . . . 134
XVI.	Number of head lettuce plants examined and found infected with aster yellows virus, Winnipeg

TABLE	PAGE
Spring crop 1963 . . . . .	135
XVII. Number of <u>Macrosteles fascifrons</u> per 50 sweeps per plot, Portage la Prairie, Summer crop 1963 . .	136
XVIII. Number of head lettuce plants examined and found infected with aster yellows virus, Portage la Prairie, Summer crop 1963. . . . .	137
XIX. Transmission of three strains of aster yellows virus by <u>Macrosteles fascifrons</u> to fourteen different hosts. . . . .	138
XX. Transmission of three strains of aster yellows virus by single <u>Macrosteles fascifrons</u> based on acquisition from fourteen different host plants . . . . .	139
XXI. Analysis of variance table for transmission of three strains of aster yellows virus "A", "B", and "C" by individual infective <u>Macrosteles fascifrons</u> to fourteen different hosts. . . . .	140
XXII. Analysis of variance table for transmission of three strains of aster yellows virus "A", "B", and "C" by individual <u>Macrosteles fascifrons</u> based on acquisition from fourteen different host plants. . . . .	141

## LIST OF FIGURES

FIGURE		PAGE
1.	Head lettuce, var. Imperial 456, showing symptoms of AYV, brown exudate on stiff, chlorotic strap-like terminal leaves . . . . .	20
2.	Cellulose nitrate cage over an aster, four-leaf stage, grown in a three-inch peat pot. . . . .	24
3.	Metacrylic plastic tube cage over an aster grown in a four-inch clay pot. . . . .	26
4.	Polyethylene bag cage over an aster grown in a three-inch peat pot. . . . .	27
5.	Greenhouse bench showing a series of polyethylene bag cages used in a transmission test. . . . .	30
6.	Aster, var. Giant Pink, plants showing symptoms of AYV strains "A", "B", and "C" . . . . .	66
7.	<u>Nicotiana rustica</u> var. <u>humilis</u> showing symptoms of AYV strains "A", "B", and "C". The plant infected with strain "B" is supported with a straw. . . . .	67
8.	Head lettuce var. Imperial 456, showing symptoms of AYV strains "A", "B", and "C" . . . . .	75
9.	Stinkweed, <u>Thlaspi arvense</u> L., rosette stage showing symptoms of AYV strains "A", "B", and "C". . . . .	76
10.	Stinkweed, <u>Thlaspi arvense</u> L., flowering stage, showing symptoms of AYV strains "A", "B", and "C". . . . .	77



FIGURE	PAGE
11. Celery, var. Utah, showing symptoms of AYV strains "B", and "C" . . . . .	78
12. Plantain, <u>Plantago major</u> L., showing symptoms of AYV strains "B" and "C" . . . . .	79
13. Barley, var. Parkland, showing symptoms of AYV in order from left to right strains "C", "A", and "B" and check. . . . .	80
14. Onion, var. Ebenezer, showing symptoms of AYV in order from left to right strains "C", "B", and "A" and check. . . . .	81
15. Flax, var. Redwood, showing symptoms of AYV strains "A", "B", and "C". . . . .	82
16. Sunflower, var. Peredovik, showing symptoms of AYV strains "A", "B", and "C" . . . . .	83
17. Sunflower, var. Commander, showing symptoms of AYV strains "A", "B", and "C" . . . . .	84
18. Carrot, var. Special Long Type Nantes, showing symptoms of AYV strains "A", "B", and "C". . . . .	85
19. Tame buckwheat, var common, showing symptoms of AYV strains "A", "B", and "C" . . . . .	86
20. Wild buckwheat, <u>Polygonum convolvulus</u> L. showing symptoms of AYV strains "A", "B", and "C". . . . .	87
21. Wheat, var. Selkirk, showing symptoms of AYV strain "B" . . . . .	88

## CHAPTER I

### INTRODUCTION

The recognition of plant virus infections as a distinct class of disorders and of insects as vectors of the plant viruses dates back to the end of the nineteenth century (Cook, 1947). Since then, it has been shown that most plant species of economic importance are infected by one or more virus diseases, many of which are transmitted by insects (Chapman, 1959). In Manitoba, wheat is known to be infected by three insect-transmitted viruses: barley yellow dwarf, aster yellows, and wheat striate mosaic; oats by two: barley yellow dwarf and oat blue dwarf; barley by four: barley yellow dwarf, oat blue dwarf, aster yellows, and wheat striate mosaic; flax by two: oat blue dwarf and aster yellows, and most other crops, vegetables and herbaceous ornamentals by at least one: aster yellows. Barley yellow dwarf, a virus disease of world distribution, was discovered in 1951 by Oswald and Houston (Bruehl, 1961), and the oat blue dwarf as recently as 1961 (Banttari and Moore, 1961).

The protection of the crops from the insect-transmitted viruses may be based on the development of crop varieties resistant to the insect or the disease, or, on the use of chemicals. Each method has its particular advantages or disadvantages, but common to each is the necessity for a thorough understanding of the vector, virus and host-plant relationships. The complexity of the relationship is illustrated by the aster yellows virus (AYV). It is a complex of virus

strains (Granados, 1965) some with different host ranges, different host effects and different vectors in different parts of its range. Macrosteles fascifrons (Stål), the six-spotted leafhopper, is the major vector, though only one of at least 25 different species of leafhoppers which are capable of transmitting AYV. In Manitoba the migrant populations of M. fascifrons are the major source, through the eggs they deposit, of the local population that develops in Manitoba. The migrants are also the major source of AYV which they carry with them and inject into the local crop and weed plants. The diseased crop and weed plants form the major source of infection for the generations of M. fascifrons which develop in Manitoba.

#### The problem

The problem is the protection of the crops from infection by AYV. The first method studied was the direct destruction of the six-spotted leafhopper by chemicals. For this purpose a series of insecticides were evaluated to determine the most effective time of application, rates and interval of application.

The second method was a study to determine the relationship of the vector and virus with a number of common host plants. The purpose was to elucidate the facts that may prove effective in reducing AYV infections, possibly through the reduction of certain host plants or in the production of resistant crop plants.

#### Importance of the problem

The host range of M. fascifrons and AYV includes most of

the cereal crops, oil seed crops, vegetables and many of the herbaceous ornamentals grown in Manitoba. Although the losses due to AYV infection seldom exceed ten per cent of most crops, these losses have practically eliminated commercial production of head lettuce in Manitoba, and constitute serious problems on carrots and celery; in some years some growers have lost their entire celery crops. The loss in most of the other crops caused by the virus disease although low, represents another addition to the cost of production and to the cost of the final food product.

#### Organization of the thesis

The chemicals used in the tests are referred to by their trade names or company designations throughout the thesis. The chemical names are given in the section on Materials and Methods Chapter III. The results of the field tests are presented in Chapters IV to VII.

The laboratory tests conducted at the Canada Department of Agriculture, Research Station, Winnipeg, to differentiate three strains of aster yellows virus, and on the ability of M. fascifrons to transmit three strains of AYV to and from fourteen different host plants are given in Chapters VIII, IX and X respectively.

Two publications based on the results presented in Chapters IV, V, VI and VII are given in Appendix A. Summary tables of data that form the basis for the final Tables presented in the text are given in Appendix B.

## CHAPTER II

### REVIEW OF LITERATURE

#### Control of insect vectors of plant viruses

The control of vectors of plant viruses to prevent the spread of diseases has been attempted with insecticides, trap crops, and various types of barriers. A survey of the literature on insecticidal control of the spread of plant viruses was presented by Broadbent (1957). In this review he listed the earlier successes and failures by a number of workers attempting to reduce the spread of spotted tomato wilt on tomatoes by thrips, potato viruses by aphids, aster yellows virus on lettuce, carrots, and endive by M. fascifrons and curly top of sugar beets by the leafhopper Circulifer tenellus Baker. Broadbent (1957) concluded from the early attempts to control the spread of viruses by using such insecticides as nicotine, derris and pyrethrum, that it is not sufficient to kill the insects on the crop, that insufficient insecticide remained active on the foliage, and very frequent applications were necessary to prevent virus spread. These were expensive and often failed because incoming insects introduced virus or spread it from plant to plant in the intervals between applications. An increased incidence of potato virus Y in potato fields sprayed with insecticides has been reported by various workers (Chapman, 1951; McEwen, 1953; Roland, 1953). McEwen (1953) showed that insecticides kept the foliage green late in the season thus making the plants susceptible to late-season infection. Chapman (1951) hypothesized that parathion increased aphid activity and

Roland (1953) suggested that it attracted aphids. According to Shanks and Chapman (1965), aphids, after initial probes made longer feeding probes on parathion-treated tobacco than on untreated leaves and vice-versa on DDT-treated leaves. Neither material affected aphid acquisition of potato virus Y but parathion tended to decrease and DDT to increase the number of transmissions to treated plants. The authors found that the insecticides required 51-180 min. to kill 90 per cent of the aphids two hours after application and much longer three days after treatment, while aphids can transmit non-persistent viruses in less than a minute. They also found that the winged aphids remain longer on parathion-treated plants than on DDT-treated plants, and, that infected aphids transmitted potato virus Y slightly more often to parathion-treated than to untreated tobacco plants.

It was shown by Becker and Rich (1956) and by Morgan (1965) that the control of aphids, Pentatrichopus spp., on strawberry beds prevented the spread of virus diseases and, as a result, increased runner and fruit production. Morgan (1965) showed that one application of Meta-Systox or dimethoate during the spring to prune trees gave a season-long control of aphids and suggested that limitation of the spread of viruses in the field through control of insect vectors, whether or not the identity and behavior characteristics of such vectors are known, is feasible.

Harrison et al (1963) and Murrant et al (1965) showed that the nematocide D-D effectively controlled two species of nematodes Xiphinema diversicaudatum (Micol) and Longiforus elongatus (de Mon) and thus

prevented the spread of virus diseases in strawberry beds. According to Murrant et al (1965) the effects lasted throughout the four year experiment.

Strong and Rawlins (1958) tested four insecticides: DDT, demeton, malathion and parathion at various dosages for the control of M. fascifrons on lettuce. The materials were sprayed on plants growing outdoors and which were taken into the laboratory for tests after different lengths of time. Their conclusion was that none of the insecticides applied as sprays was very effective for as long as three days. Further tests were carried out by Strong and Rawlins (1958) in the field using the same four insecticides and using populations of infective insects. The authors were able to differentiate between incidence of disease on sprayed and unsprayed lettuce, and in the residual effectiveness of the insecticides tested. The differences between the insecticides were not evident and the degree of protection was not satisfactory for commercial conditions. Chiykowski (1958), found malathion effective for the control of M. fascifrons on head lettuce and prevention of infection by AYV. Miller (1960) recommended DDT and malathion for the control of M. fascifrons on carrots and lettuce in Ontario.

Thompson and Rawlins (1961) tested the systemic insecticides phorate, dimethoate and Di-Syston to control M. fascifrons and reduce the incidence of lettuce yellows. The insecticides, applied at the rate of one pound per acre at time of seeding as granules or as drenches, were more effective than seed treatments and equally or more effective than malathion applied twice weekly as a spray. Phorate was more