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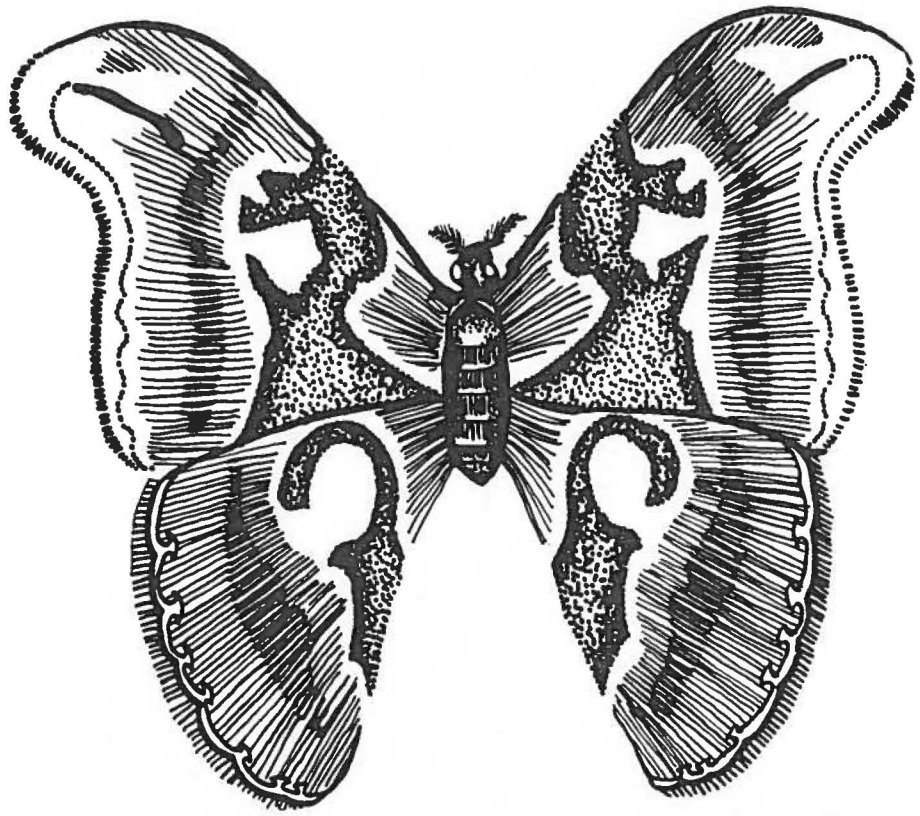

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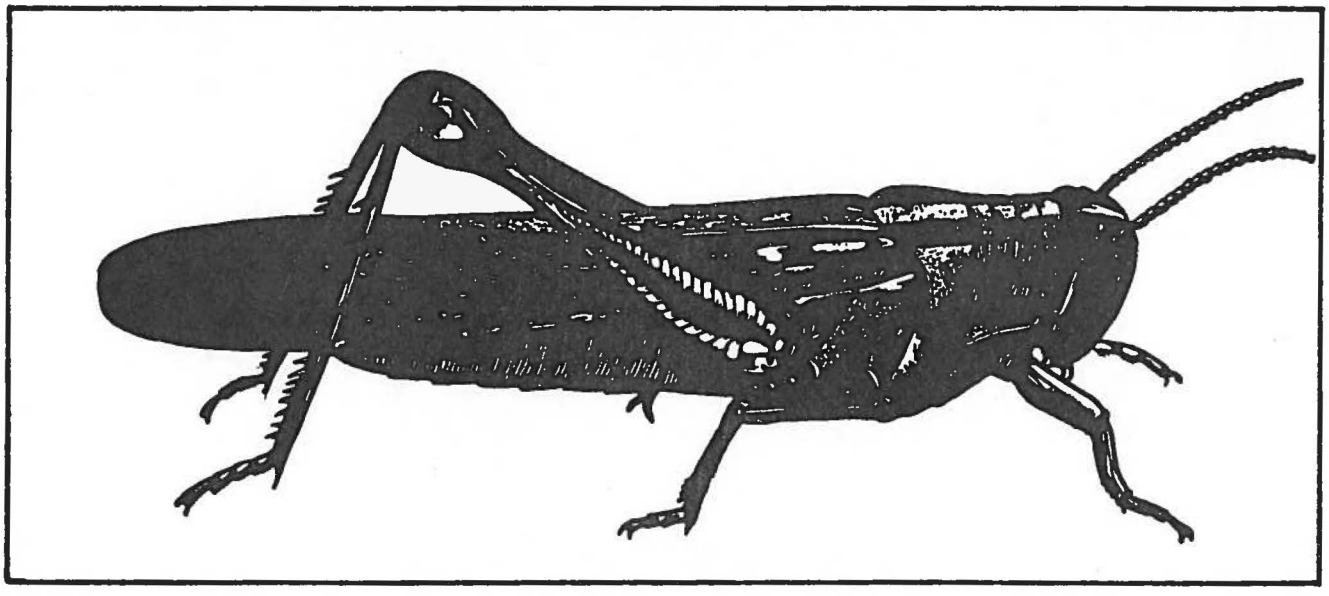

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An official publication of the Entomological Society of Manitoba, an organization to foster the advancement, exchange and dissemination of entomological knowledge

CONTENTS

USE OF REGRESSION ANALYSIS TO DETERMINE DOSAGES OF HYDROGEN PHOSPHIDE APPLIED TO STORED GRAIN[1]

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ABSTRACT: Comparison of PH_3 concentrations in perforated and non-perforated steel drums showed that it was possible to verify applied dosages of PH_3 by intercepts obtained from regressions of $\log_{10}$ of gas concentrations on time. Analysis of data on gas concentration in grain that was fumigated in concrete bins for commercial purposes showed that there is close agreement between calculated dosages and actual amounts applied. A high correlation coefficient was obtained for steel drums and filled, and partly filled concrete elevator bins ($r = -0.87$ to -0.99) when gas concentration data were transformed to $\log_{10}$ scale and correlated to time.

INTRODUCTION

Phosphine gas (PH_3) generated from tablets and pellets of aluminum phosphide is often used to control infestations in stored grain (Schesser 1967; Wainman *et al.* 1974). The tablets or pellets may be dispersed through pipes inserted in the grain or, alternatively, may be applied by hand or by means of automatic applicators as the grain is binned (Heseltine 1973). Where large numbers of tablets or pellets are used, it is often desirable to verify the amount of gas generated after application to insure that the operator has applied sufficient tablets or pellets to kill the target species (Cogburn 1974; Dumas and Monro 1966).

During routine monitoring of gas concentrations in grain in steel drums (Barker 1974a), it became evident that there was a relatively rapid rise in PH_3 concentrations for one to 3 days after treatment followed by a gradual decline. Some of the probable causes for the decline of PH_3 concentrations include diffusion of the gas out of the bins (Barker 1974b), effects of wind (Mulhearn *et al.* 1976) and breakdown of the molecule (Tkachuk 1972). Experiments were conducted with metal drums and with 2 concrete elevator bins which contained wheat, to establish if it was possible to use the data from gas concentration measurements to calculate the amounts of hydrogen phosphide applied to the drums and concrete bins at the start of fumigation.

MATERIALS AND METHODS

Six steel drums, 1.76 m high and 0.57 m in diameter, each with a capacity of 454 liters were used. Each of 3 of the drums had 2 small holes, 2 mm in diameter, at 0.09 and 1.67 m from the bottom; the other 3 drums had no holes.

Six aluminum phosphide pellets (Degesch, Frankfurt am Main, Federal Republic of Germany) were placed in each of the 6 drums. A piece of water-soaked paper towelling, 1.20 m long and 0.20 m wide was attached to the inside wall of each drum to increase humidity and facilitate generation of PH_3. The drums were then sealed by applying self-adhering aluminum tape (Aluminum Foil Tape, Nashua Corporation, Chicago, Ill.) to secure the lids.

PH_3 concentrations were measured with Drager (Dragerwerk AG, Lubeck, Federal Republic of Germany) tubes at various times after initiation of the experiment (Table 1). The Drager tubes were attached to a 0.15 m steel hypodermic needle by means of a 0.04 m length of rubber tubing. The needle was pushed through self-adhering aluminum

[1] Contribution No. 765, Agriculture Canada, Research Station, Winnipeg, Manitoba.

tape over small holes which had been drilled 1.02 m from the bottom of each drum. The drums were placed outdoors for 108 h. Temperatures in the shade ranged from 17 to 34°C.

Two large concrete bins were also used. One had a capacity of 136 metric tons (5,000 bu U.S.) and contained 54 metric tons (2,000 bu U.S.) of wheat. The other bin had a capacity of 81 metric tons (3,000 bu U.S.) and contained 79 metric tons (2,936 bu U.S.) of wheat. The first mentioned bin received 1,100 aluminum phosphide pellets and the second received 800 pellets. The pellets were placed in the grain streams as they entered the bins. Gas concentrations were measured in the head spaces above the grain at various times during fumigation (Table 2). The grain temperature in both bins was 18.3°C.

Least squares regression and correlation analyses were performed on the untransformed data obtained from the drums and from the elevator bins. The analyses were repeated after the concentrations of gas (ppm) were transformed to a $\log_{10}$ scale (Snedecor and Cochran 1974).

RESULTS AND DISCUSSION

The average PH_3 concentrations found in the steel drums declined continuously from the first reading made 29 h after application of the pellets (Table 1) and were lower in the perforated than in the nonperforated drums. Similar patterns of gas loss were also observed in the two large grain bins (Table 2).

The difference in gas concentrations between perforated and nonperforated drums (Table 1) increased up to 60 h, after which the difference diminished steadily ($Y = 394.97 - 2.111X$; $r = -0.9723$), and gas concentrations decreased more rapidly from the nonperforated than from the perforated ones. PH_3 concentrations dropped below 100 ppm in 90 h for perforated drums and in 144 h for nonperforated drums. The gas concentrations in the elevator bins (Table 2) were different, reflecting differences of size and rates of gas loss.

Table 1. Average concentrations of PH_3 in 454-liter steel drums at various times during fumigation.

	PH_3 (ppm)		
Hours after application	Perforated drums	Nonperforated drums	Difference
29.25	660	863	203
34.75	610	826	216
46.75	510	720	210
49.25	525	758	233
53.25	381	628	247
59.75	280	551	271
70.58	206	466	260
73.75	186	421	235
79.15	143	331	188
83.75	105	333	228
94.75	76	296	220
98.00	76	270	194
104.50	45	231	186
121.25	25	135	110
147.75	16	90	74
159.83	6	72	66
167.75	19	65	46

Table 2. PH_3 concentrations in elevator bins at various times during fumigation.

136 metric ton bin		81 metric ton bin	
Hours after application	PH_3 (ppm)	Hours after application	PH_3 (ppm)
168	650	70.00	350
192	700	88.25	230
216	650	95.50	175
240	550	112.25	100
264	500	119.50	90
		136.50	55
		142.50	35
		160.25	30

Gas loss in the perforated drums was expected because of diffusion of the gas through the holes, because the daily rise and fall of temperatures and barometric pressures pumped the gas out of the drums (Barker 1974b) and because of wind effects (Mulhearn *et al.* 1976).

The fact that the nonperforated drums lost gas throughout the experiment suggests chemical breakdown of the PH_3, as suggested by Tkachuck (1972), or its sorption onto the sides of the drums or onto paper towelling. In practical fumigation, a reduction of gas concentration occurs regardless of the airtightness of the grain bins.

Tkachuk (1972) showed that with a 120 h exposure to grain to $^{32}PH_3$ in 0.075 liter glass bottles, 23 to 44% of the ^{32}P could be recovered from the inorganic residues and confirmed the findings of Robinson and Bond (1970) that PH_3 forms nonvolatile inorganic residues.

The correlation coefficients obtained from the untransformed data were −0.8595 and −0.9297 for the perforated and nonperforated drums, respectively, and −0.866 and −0.9421 for the 136 and 81 metric ton elevator bins, respectively. When the measured gas concentrations were transformed to a $\log_{10}$ scale, the correlation coefficients were improved to −0.9762 and −0.9966 for the perforated and nonperforated drums, respectively, and to −0.8729 and −0.9922 for the 136 and 81 metric ton elevator bins, respectively (Table 3). Thus, gas concentrations were best related to time when the gas concentrations were transformed to a $\log_{10}$ scale.

If it is assumed that the gas loss is constant (on a $\log_{10}$ of ppm scale) from the start of the experiment, the antilog_{10} of the intercepts from the regression equations of the transformed gas concentrations on time should correspond to the calculated maximum gas concentrations in the drums and bins. The gas concentrations used for this analysis should be the ones obtained after the peak concentration has been reached. The assumption that the antilog_{10} of the intercepts and the dosages applied should be the same was verified because the calculated concentrations of PH_3 and the antilog_{10} of the intercepts were about the same for both kinds of drums and for both bins (Table 3).

For the perforated steel drum, the antilog_{10} of the intercept indicated that 1,960 ppm of PH_3 had been applied, about 3.4% more than the 1,897 ppm actually placed in the drum. The results for the nonperforated steel drum showed a calculated 1,850 ppm which was 2.3% less than the 1,897 ppm actually applied.

For the 136 metric ton bin, the antilog_{10} of the intercept indicated that a maximum calculated concentration of 1,206 ppm should have been obtained at the start of the experiment, very close (within 2.3%) to the actual concentration of 1,179 ppm (Table 3) obtained from the 220 g PH_3 (= 1,100 pellets of A1P) diluted into the airspace of the bin. Similarly, for the 81 metric ton bin, the antilog_{10} of the intercept indicated that 2,753 ppm PH_3 should have been obtained at the start of the experiment, close (within 8.0%) to the actual 2,550 ppm obtained from diluting 160 g PH_3 (= 800 pellets A1P) into the

Table 3. Calculation of applied dosages of PH_3 in steel drums and two elevator bins, and the regression analyses of the data.

	Drums		Elevator bins	
	Perforated	Non-perforated	136 ton	81 ton
Dosages (ppm)[1]				
Actual	1897	1897	1179[2]	2550[2]
Calculated	1960	1850	1206	2753
Regression Analyses				
Slope	−0.0143	−0.0088	−0.0013	−0.0126
Intercept	3.2922	3.2671	3.0814	3.4398
$S_{y.x}$	0.1387	0.0318	0.0339	0.0517
S_b	0.0008	0.0002	0.0004	0.0006
r	−0.9762	−0.9966	−0.8728	−0.9922
t	17.44	6.88	3.09	19.48
n	17	17	5	8

[1] Assume 1 g PH_3/m^3 = 718 ppm (Monro 1969).
[2] Assume 40% air space in the wheat.

airspace of the bin. These trials constituted a further verification of the assumption that the $antilog_{10}$ of the intercept from the regression is similar to the calculated concentration. Some of the small discrepancies between $antilog_{10}$ of the intercept and the calculated concentration were probably due to the variation of the per cent airspace in the grain, to errors in the measurement of the internal volume of the bins, or possibly to an erroneous count of the number of pellets applied.

These experiments have shown that there was a very good negative correlation between the log_{10} of declining gas concentrations (ppm), in the steel drums and elevator bins, and time (h). Furthermore, it was shown that the $antilog_{10}$ of the intercepts obtained was very close to the actual amounts of PH_3 applied to the drums and bins. This relationship may be very useful to monitoring agencies, for verification of the amounts of PH_3 applied to elevator bins for commercial control of infestations.

ACKNOWLEDGEMENTS

I thank Mr. John Elvidge of the Canadian Grain Commission, Vancouver, B.C. for the data from the 136 metric ton bin, and Mr. D. Kurtz for his technical assistance.

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(Received 10 November, 1977)

THE OUTBREAK OF BERTHA ARMYWORM *MAMESTRA CONFIGURATA* (NOCTUIDAE: LEPIDOPTERA), IN ALBERTA, 1971 TO 1975[1]

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ABSTRACT: During the outbreak of bertha armyworm, *Mamestra configurata* Walker, in Alberta, from 1971 to 1974, methods of predicting infestations were quickly developed, put into operation, and modified as dictated by experience. Light traps for adults, pupal sampling and sampling of early larval instars were used to estimate and predict the abundance of bertha armyworm. Analysis of these techniques and their usefulness in predicting damaging infestations showed that although the predictions were of considerable value, the techniques needed refinement to increase accuracy and decrease cost. The techniques used to estimate bertha armyworm abundance also provided information on the life history and phenology of the bertha armyworm.

INTRODUCTION

The outbreak of bertha armyworm, *Mamestra configurata* (Walker), of 1971 to 1974, affected a high proportion of the fields of rape, *Brassica napus* L. and *B. campestris* L., across the prairie provinces. Although an increase in numbers of bertha armyworms was noted in 1970, the severity of the outbreak in 1971 was not predicted and emergency measures were required to register and obtain an effective insecticide, to organize its distribution, and to allocate spray planes to the affected areas. During the course of the outbreak, it became obvious that a system of monitoring populations was needed to provide an early identification of areas of potential infestation, followed by surveillance of these areas up to the time of decision on the need for insecticide application.

The outbreak ended before these techniques could be adequately tested. However, an evaluation of the available results is needed to provide a basis for planning a program for future outbreaks. This evaluation, plus information on the bertha armyworm infestations, life history and phenology are presented in this paper.

METHODS

In 1972, personnel of the Plant Protection Laboratory, Alberta Department of Agriculture, initiated a number of sampling programs designed to obtain the information needed for extension and control operations. These programs were expanded and refined, on the basis of accumulated experience, as the outbreak progressed. Soil sampling for pupae, during the fall, was conducted to provide an estimate of the area where there were populations with a potential for damaging the crop in the next year. This estimate, it was anticipated, would provide the pesticide industry with sufficient information and time to ensure the availability of insecticides. The following spring, pupal sampling was designed to refine the fall estimate. Soil samples were collected in 51 fields in the fall of 1972 and 56 fields in the spring of 1973. The number of fields sampled was reduced in the fall of

[1] Contribution No. 823, Agriculture Canada, Research Station, Winnipeg.

1973 and the spring of 1974 and then the program was discontinued because populations of bertha armyworm were low.

The numbers of bertha armyworm adults caught in light traps were used to further define areas with potentially damaging infestations to allow government agricultural field staff to plan and execute extension activities (local surveys, organization of spray programs, talks, etc.). The number of light traps was 3 in 1972, 13 in 1973 and 17 in 1974.

A limited survey, to detect the presence of small larvae of the bertha armyworm, was conducted in 1972 and expanded in 1973. The results of these surveys were used to confirm the presence and time of occurrence of potentially damaging infestations so that producers could be advised to examine their fields. In 1972 and 1973, a maximum search time of 10 minutes per field was used but variation between individuals in their rate of searching was found to cause considerable variation in the number of sampling units examined. In 1974, a maximum of 6 sample units was searched in each field sampled.

The sampling techniques also provided information on the life history and phenology of the bertha armyworm. These data have been supplemented by information collected in Manitoba. Additional information on the course of the outbreak was used to place the 1971 to 1974 outbreak within the history of bertha armyworm infestations. Specific details of the techniques used are presented with the results.

RESULTS

Life History and Phenology

The bertha armyworm completes one generation each year in the prairie provinces and appears capable of overwintering only in the pupal stage. Pupation usually occurs in late August, about the time that rape crops are swathed. In different years and localities pupation may occur from late July to late September. In some years, adults of a second generation emerge in late August or September, but offspring from this generation do not survive the winter.

In the spring, emergence of bertha armyworm adults, as determined by emergence traps located near Wainwright, Alberta (52° 49'N, 110° 52'W), in 1972, Neepawa, Manitoba (50° 15'N, 99° 40'W), in 1973, and Bowsman, Manitoba (52° 14'N, 101° 27'W), in 1974, began from 8 June to 7 July and continued for 3 to 5 weeks (Figure 1). Catches of moths in black-light traps operated in the vicinity of these emergence traps reflected the emergence pattern, with some variability attributable to the relative location of the two types of traps. At Bowsman, where the light traps were located adjacent to the fields containing the emergence traps, the patterms nearly coincided, whereas at Neepawa, where the light traps were located adjacent to new rape fields a few miles from the emergence traps, the patterns were similar, but the light trap data indicated moth activities beginning and ending about 7 days later than moth emergence. At Wainwright, the light trap was adjacent to the field containing the emergence traps and the patterns of catch coincided well until 23 June. During the period 24 June to 2 July, no moths were collected in the light traps and the second period of moth catches seems to reflect the activity of moths from other fields.

Additional data from black-light traps operated by the Plant Industry Laboratory, Alberta Department of Agriculture, show considerable variation in the dates of moth activity (Table 1). The extremes were 15 June and 24 August. The date on which 50% of the total catch accumulated varied from 30 June to 17 July.

In the laboratory, most female moths begin to oviposit within 5 days of emergence. There are few field observations on eggs. Collections made in fields near Neepawa, Manitoba, in 1973, showed that egg laying occurred from 4 to 23 July, with a peak from 13 to 18 July. The earliest and peak dates of egg laying were 7-10 days later than corresponding dates from light trap catches shown in Figure 1.

In Alberta, the results of early larval surveys indicated that hatching was completed by mid to late July in 1973 and from late July to early August in 1974 (Table 2). In 1972, eggs and larvae in the first three instars were found throughout the rape growing areas of

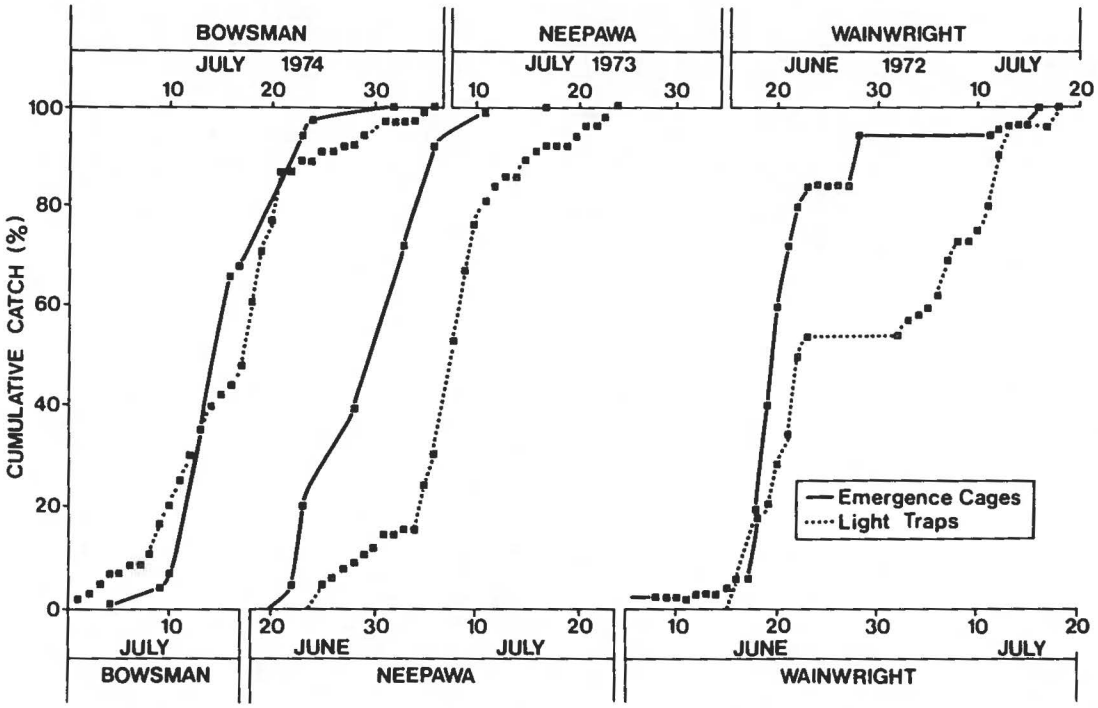

Figure 1. Cumulative catches (%) of moths in emergence traps and nearby black light traps near Bowsman and Neepawa, Manitoba and Wainwright, Alberta.

Table 1. Dates of first, last and 50% capture of bertha armyworm moths in light traps in Alberta, 1972 to 1975. Town(s) nearest to light traps in parentheses.

Location	Year	Number of traps	Mean number of moths per trap	Dates of moth capture		
				First	50%	Last
Crop District 2						
(Brooks, Nobleford,	1973	3	152	15 June	10 July	31 July
Three Hills,	1974	3	17	30 "	14 "	24 Aug.
Strathmore, Vulcan)	1975	5	3	26 "	11 "	23 July
Crop District 4	1972	2	146	18 "	30 June	18 "
(Killam, Vermilion,	1973	3	72	15 "	4 July	30 "
Wainwright,	1974	3	3	4 July	13 "	19 "
Stettler)	1975	3	0	–	–	–
Crop District 5	1972	1	30	9 July	11 July	16 July
(Bon Accord,	1973	3	88	18 June	9 "	30 "
Coronado, Legal,	1974	1	41	26 "	12 "	4 Aug.
Lacombe, Warburg)	1975	2	0	–	–	–
Crop District 7						
(Deadwood, DeBolt,	1973	4	18	23 June	9 July	27 July
Culp, Grande Prairie,	1974	4	10	26 "	12 "	4 Aug.
Fairview, Valhalla Centre)	1975	5	0	–	–	–

Table 2. Occurrence of bertha armyworm egg masses (expressed as a percentage of the total number of recorded occurrences of egg masses and larvae) by date of sampling during surveys for young larvae of bertha armyworm in 1973 and 1974.

	Crop district(s)			
Date	1, 2, 3	4, 5	6	7
1973				
July 16		27 (22)		
17	18 (11)[1]	6 (18)	0 (2)	
18	5 (22)	17 (6)	0 (3)	
19	0 (2)	11 (9)	0 (1)	
20		6 (17)	0 (3)	
23-25				0 (35)
1974				
July 23	20 (5)	0 (1)		
24-27	0 (6)	0 (4)		
28		20 (5)		
July 31, August 1		0 (8)	25 (4)	
August 6				50 (2)
7				20 (10)
8				8 (12)
9				0 (5)

[1] Number of observations in parentheses.

Alberta, excluding the Peace River District, during surveys from 17-20 July. Peak egg hatching would have occurred during the last week of July.

History of Infestation

Infestations of bertha armyworm occurred on various crops in 1921-22, 1927-30, 1940-44, 1947-48, 1953-56, 1962-63, and 1966-68 (King 1928, 1929; Durksen 1971). The early infestations were on flax, alfalfa, sweet clover and assorted vegetables and flowers, but since 1940 rape has been the main crop attacked. As previous infestations on rape had not been serious, reports of widespread but light infestations in 1970 did not cause alarm. However, in 1971, an outbreak of unprecedented severity occurred across an area roughly 50 miles wide, extending from Edmonton in Alberta to Swan River and Dauphin in Manitoba.

In Alberta, the 1971 infestation occurred in the east-central parklands, mainly in Crop District 4 between the North Saskatchewan and Battle rivers and from Edmonton east to the Saskatchewan boundary (Figure 2). The infestation area spread, in 1972, to include most of Alberta's rapeseed growing areas (Shields and Ferguson 1975), decreased in 1973 and was limited to a few areas in 1974. The acreage sprayed with insecticides in crop districts (Table 3) shows an annual trend similar to that of the infestation map (Figure 2). However, when the acreage sprayed was mapped, a somewhat different pattern of infestation was revealed (Figures 3, 4, 5). The area of infestation, based on the acreage sprayed tended to be underestimated because fields with subeconomic densities, fields in which infestations were detected too late to spray, and fields in which nearly mature crops were harvested rather than sprayed, were omitted.

Pupal Sampling

The number of bertha armyworm pupae in a field was estimated by examining the soil from 10 samples, each 0.84 m^2 (1 yd^2), to a maximum depth of 25 cm. Shallower

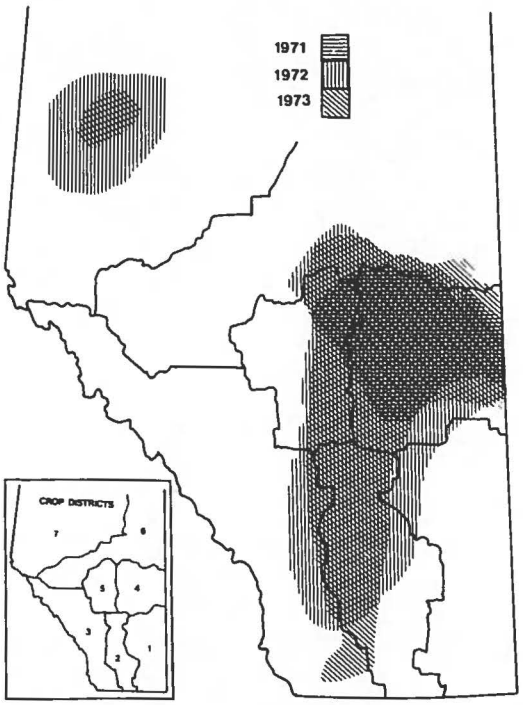

Figure 2. Areas of Alberta infested by bertha armyworm, 1971 to 1973.

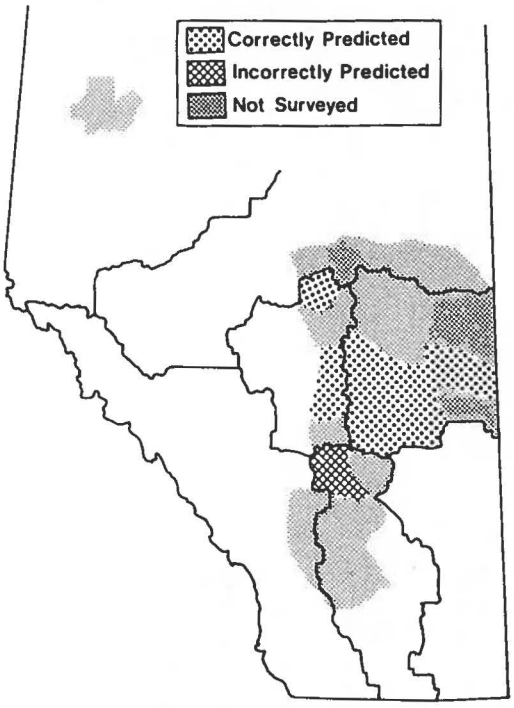

Figure 3. Areas of Alberta treated with insecticides against bertha armyworm larvae in 1972, showing those areas in which the outbreak was predicted on the basis of the early larval survey ($\geqslant 50\%$ of the fields infested in a rural municipality) and those areas in which no outbreak was predicted ($< 50\%$ of the fields infested).

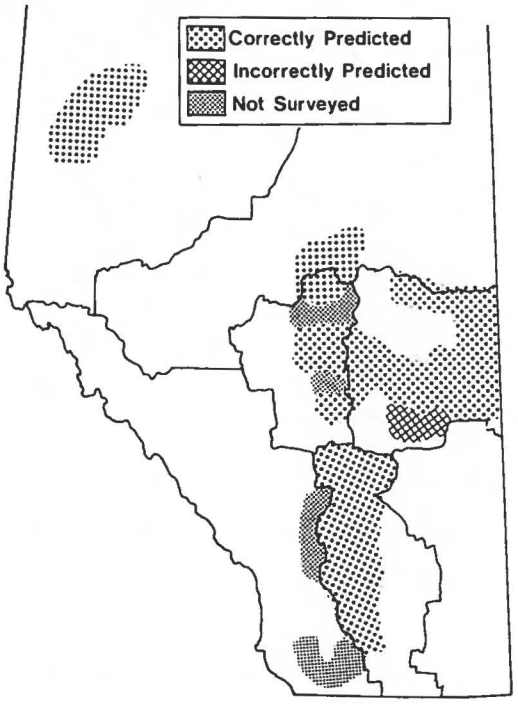

Figure 4. Areas of Alberta treated with insecticides against bertha armyworm larvae in 1973, showing those areas in which treatment was correctly or incorrectly predicted on the basis of the percentage of fields infested and the density index for each rural municipality.

Figure 5. Areas of Alberta treated with insecticides against bertha armyworm larvae in 1974, showing those areas in which treatment was correctly or incorrectly predicted on the basis of the percentage of fields infested and the density index for each rural municipality.

Table 3. Number of acres of rape sprayed[1] with insecticides for the control of bertha armyworm by crop district in Alberta, 1971 to 1974.

Crop district	1971	1972	1973	1974
1	0	0	0	0
2	12,541	30,649	16,637	100
3	1,600	4,250	2,950	0
Subtotal	14,171[2]	34,889 (11.8)[3]	19,587 (7.8)	100 (0.1)
4	162,936	144,834	3,752	150
5	4,539	21,879	16,874	2,120
6	900	12,119	5,263	0
Subtotal	168,375[2]	178,832 (30.8)	25,889 (5.3)	2,270 (0.5)
7	0	12,185 (2.9)	9,620 (1.7)	500 (0.1)
Total	182,516 (9.2)	225,906 (17.4)	55,096 (4.2)	2,870 (0.2)

[1] Data provided by Alberta Agricultural Service Board; seeded acreage taken from Prairie Grain Variety Survey, published by Canadian Cooperative Seed Growers.

[2] Information on seeded acreage by crop district not available.

[3] Area sprayed as a percentage of the total area seeded to rape in parentheses.

samples were taken if compacted soil was reached in less than 25 cm. The 10 sample units were taken 30 m apart along a line at a right angle to the field border. The soil from each sample was sifted (6 mm mesh) and the living pupae counted.

Samples were taken from unsprayed fields and from fields that had been sprayed with insecticide to control bertha armyworm larvae. Comparison of pupal abundance in sprayed and unsprayed fields in the same areas, using a series of "t-tests", showed no significant differences ($P > .05$) so the spraying history was disregarded in subsequent analyses.

Pupal abundance was determined by fall and spring surveys for the 1972-73 and the 1973-74 bertha armyworm generations. In most cases, the same fields were sampled in both the fall and the spring. The mean number of pupae /0.84 m^2 was calculated for each of the three major rape-growing areas of Alberta: Crop Districts 1-3, Crop Districts 4-6 and Crop District 7 (Table 4). Variability in pupal density was high: the standard deviation was approximately twice the mean at the higher densities and six times the mean at the lowest density. Statistical studies using these and other data (W. J. Turnock, unpublished data) on bertha armyworm pupal sampling indicate that, for 10 sample units of 0.84 m^2 per field, the probability that the true mean differs from the estimated mean by more than ± 50% of the estimated mean decreases with increasing mean density. Thus, at an estimated mean density of .22 pupae/0.84 m^2, there is a 41% chance that the true density is outside the ± 50% error limits (.11-.33); at .99 pupae/0.84 m^2 there is a 23% chance of exceeding the 50% error limits (.49-1.49) while at 1.99/0.84 m^2 the chance is 20% of exceeding the 50% limits (.99-2.99).

The number of fields that were sampled per district was limited by available manpower and suitable weather. The high variability of the estimates suggests that more fields should be examined, but the high cost and low efficiency of the sampling techniques make such an increased effort unlikely. A sequential sampling system, using the most efficient size of sampling unit, is needed.

The potential larval population of an area, the following summer was rated as follows on the basis of the pupal densities: $\leqslant$1/0.84 m^2 = light infestation; $>$1/0.84 m^2 = moderate to severe infestation. The predicted infestations for 1973 and 1974 are shown in Figures 6 and 7.

The 1972 fall pupal survey over-estimated the area of infestation, as measured by the application of insecticide (Figure 6). Insecticides were applied in three areas outside the area of predicted infestation. All of these areas were adjacent to areas of predicted infestation; the one in the southern part of Crop District 3 was not included in the pupal survey. In 1974, the area treated with insecticide exceeded that predicted by the fall pupal survey (Figure 7). The fall pupal survey provided only a very crude estimate of the area of infestation.

The 1973 spring pupal survey showed that overwintering mortality had reduced pupal populations to a level of less than 1/0.84 m^2 (Table 4) and could have been used to improve the prediction of the area of infestation. However, the improvement was slight for the effort needed to collect the information. The 1974 spring pupal survey did not improve the prediction of the 1974 infestation.

Adult Sampling

The abundance of bertha armyworm moths has been monitored in Alberta since 1972 using New Jersey light traps (Mulhern 1942) modified by replacing the incandescent bulb with a BLB $F_{15}T_8$ fluorescent tube mounted horizontally. The traps were located on the farmsteads of cooperating farmers who emptied the traps daily or twice weekly from early June to 31 July, in 1972 and 1973, and from 1 May to 31 August, in 1974 and 1975. The collections were shipped to Edmonton where staff of the Entomology Section, Plant Industry Laboratory, identified and counted the bertha armyworm moths.

The number of traps was increased from 3 in 1972 to 17 in 1975. These traps were located throughout the agricultural areas of Alberta and from 0-205 adults/trap/season were collected. The mean number of adults per trap, by crop districts (Table 2), provided a reasonably good picture of the progress of the outbreak as shown by Figure 2, and the

Table 4. Mean numbers and standard deviation (in parentheses) of pupae/0.84 m^2 and the proportion of the fields sampled with mean numbers > 1.0/0.84 m^2 in Alberta, 1972 to 1974.

Generation and time of sampling	Crop districts 1, 2, 3			Crop districts 4, 5, 6			Crop district 7		
	N fields	Pupae/ 0.84 m^2	p > 1.0/ 0.84 m^2	N fields	Pupae/ 0.84 m^2	p > 1.0/ 0.84 m^2	N fields	Pupae/ 0.84 m^2	p > 1.0/ 0.84 m^2
1972-73									
Fall	8	5.0 (6.4)	1.0	35	5.9 (12.0)	.51	8	3.3 (6.6)	.88
Spring	8	1.9 (2.8)	.88	40	2.5 (6.4)	.40	8	2.0 (4.7)	.75
1973-74									
Fall	9	0.4 (0.9)	.11	15	0.3 (0.6)	0	12	0.7 (1.6)[1]	.33[1]
Spring	7	0.1 (0.6)[2]	0	2	1.0 (2.1)[2]	.50	6	2.4 (3.8)[2]	.50[2]

[1] Pupation not complete in some fields at time of sampling.

[2] Fields with low pupal densities in the fall survey were omitted from the spring survey.

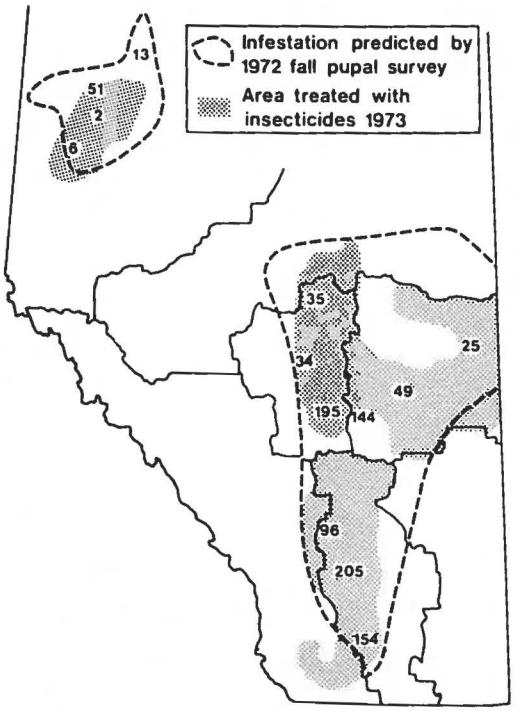

Figure 6. Area of infestation by the bertha armyworm predicted for 1973 on the basis of the 1972 pupal survey, and the area treated with insecticide against bertha armyworm larvae in 1973. Numbers of bertha armyworm moths caught in light traps in 1973 also shown.

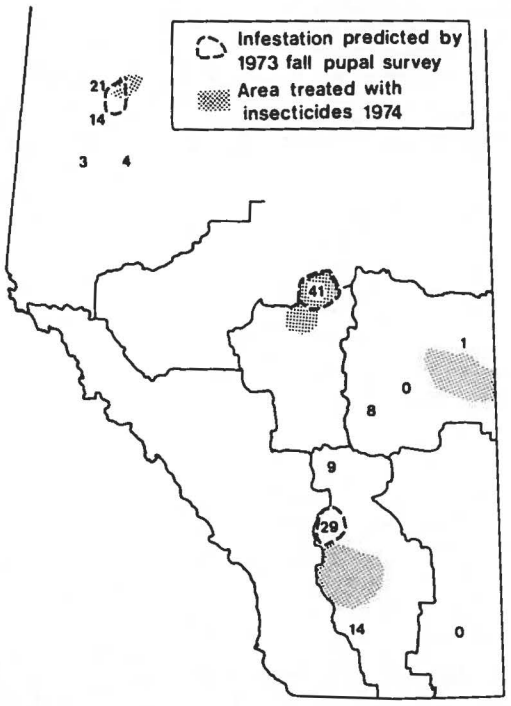

Figure 7. Area of infestation by the bertha armyworm predicted for 1974 on the basis of the 1973 fall survey, and the area treated with insecticide against bertha armyworm larvae in 1974. Numbers of bertha armyworm moths caught in light traps in 1974 also shown.

individual trap collections in 1973 agreed fairly well with the predictions based on the fall pupal survey (Figure 6). The trap collections in 1974 did not agree with estimates based on the pupal survey of the previous fall (Figure 7).

The catch of adult bertha armyworm in light traps did not, in general, provide a useful prediction of larval outbreaks (Figures 6, 7). In rural municipalities with light traps, the mean catch was 74 ± 72 moths (range 2-205) for those in which spraying was subsequently carried out and 11 ± 9 (range 0-29) for those in which there was no spraying. A more intensive system of light traps might improve the predictive value but the heavy workload in sorting and identifying light trap samples makes this approach unworkable. However, a relatively small number of traps could provide information on the phenology of bertha armyworm on which the larval surveys could be timed more accurately. The development of a specific sex attractant for bertha armyworm (Struble *et al.* 1975; Chisholm *el al.* 1975) offers the promise of an adult trapping method which avoids nearly all the problems of sorting and identifying bertha armyworm. Information will have to be gathered on the number of traps needed to estimate adult numbers and the relation between these numbers and subsequent larval outbreaks.

Larval Sampling

Surveys to determine the extent and severity of bertha armyworm infestations, based on the occurrence of early larval instars, were conducted from 1972 to 1974. The results were used to alert farmers of the need to examine their fields before damage became serious.

Sampling teams of 2 or 3 men examined fields in crop districts where infestations were expected on the basis of light-trap catches or the presence of infestations the previous year. Members of a team entered each field about 15 m apart, penetrated the field about 10 m and searched for bertha armyworm eggs and larvae. The sampling unit consisted of the plants and ground surface that could be carefully searched from a squatting or kneeling position without turning the body, an area of about 0.5 m^2. If no bertha armyworm larvae were found, additional searches were made at about 10 m intervals along lines perpendicular to the field edge. In 1972 and 1973, sampling stopped when a larva was found or after 10 minutes had elapsed. Different individuals were found to search at very different rates. In 1974, the method was changed so that sampling was halted if no larvae were found in 6 sampling units. In 1972, only the number of larvae was recorded: in 1973, the time elapsed to finding the first larvae and, in some cases, the stage of insect development was recorded. In 1974, the number of larvae, their stage of development and the number of units sampled were recorded.

The ideal timing for early larval surveys would be when the larvae are in the first and second instars. If the survey is too early, infestations would be underestimated because eggs are more difficult to find than larvae. If too late, the lead time for alerting the growers would be reduced. The 1973 and 1974 surveys were slightly too early in most crop districts, as eggs were found in some of the fields (Table 3).

In 1972, a limited survey included 63 fields in 8 rural municipalities. In 1973 and 1974, the survey was extended to include most of the rape-growing regions of Alberta. Two hundred and twenty fields in 37 rural municipalities were examined in 1973 and 221 fields in 36 rural municipalities in 1974. The surveys in 1973 and 1974 were completed in 13 to 15 working days, with an average of 16 fields examined per day.

The percentage of fields infested (FI) with bertha armyworm was calculated for each rural municipality sampled in 1972 to 1974. For each municipality sampled in 1973 and 1974, a second measure of bertha armyworm abundance, a density index (DI), was calculated:

$$DI = \left(\frac{M+1-S_i}{M}\right) \frac{100}{F}$$

where M = the maximum duration of sampling effort (10 minutes in 1973, 6 sample units in 1974); S_i = the time elapsed (1973) or the number of units sampled (1974) to find larvae in the i^{th} field; and F = the number of fields in which larvae were found. This DI ranged from 0, where no larvae were found, to 100 where larvae were found in the first sample in every field.

The percentage of fields infested, the density index and various combinations of the two measures, were examined as predictors of the pattern of use for insecticidal treatment. Predictions based on several different thresholds were made and compared to the use pattern of insecticides for each rural municipality. For 1972, when only FI could be calculated, the best predictor of rural minicipalities in which insecticidal treatment was used was FI $\geqslant$ 67 or DI $\geqslant$ 50 (Table 5). In each of 1973 and 1974, the need for treatment was correctly predicted for all but one of the municipalities surveyed (Figures 4, 5). The 1973 exception, County 18, where 300 acres were treated, was based on a survey of a single field in which no larvae were found (FI = FD = O). In 1974, the exception was Municipal District 61, with 150 acres treated, where the prediction (FI = DI = O) was based on 3 fields. The sampling method used in 1974, when a maximum of 6 sample units were taken, gave better predictions than the 1973 method using a time maximum.

The results of larval sampling, expressed in terms of the percentage of fields infested and a density index, shows promise as a method of identifying rural municipalities in which control measures would be required. The effectiveness of the larval survey in 1972 to 1974 was reduced, in some areas, by poor timing and by an uneven distribution of the fields surveyed (i.e. the number of fields sampled varied from 1-16 per rural municipality). The timing could be improved by predicting larval occurrence from adult flight or by the accumulation of heat units as described by Gage *et al.* (1976).

Table 5. Percentages of correct predictions of rural municipalities needing insecticidal control of bertha armyworm, and percentages of correct predictions of acreage sprayed, based on different threshold values of the percentage of fields infested (FI) and density index (DI)[1], Province of Alberta 1972 to 1974.

Threshold[1]		Insecticidal sprays applied						Insecticidal sprays not applied	
		1972		1973		1974		1973	1974
FI	DI	Units[2]	Acreage	Units[2]	Acreage	Units[2]	Acreage	Units[2]	Units[2]
⩾67	–	75	81	50	49	0	0	36	91
⩾50	–	88	90	91	95	50	91	21	69
–	⩾67	–	–	61	72	25	4	36	81
–	⩾50	–	–	87	98	75	95	7	53
⩾67	⩾67	–	–	74	83	75	95	21	75
⩾67	⩾50	–	–	96	99	75	95	0	50
⩾50	⩾67	–	–	91	95	75	95	7	62
⩾50	⩾50	–	–	96	99	75	95	0	50
		8[3]	111,763[4]	23[3]	49,623[4]	4[3]	2,750[4]	14[3]	32[3]

[1] DI and FI calculated from records of early larval survey in Alberta, 1972 to 1974.

[2] Unit = rural municipality.

[3] Number of observations.

[4] Total acreage sprayed in the rural municipalities.

DISCUSSION

The methods used in Alberta from 1972 to 1974 to predict the location and intensity of bertha armyworm infestations were useful as guides for the provision of insecticides, the allocation of spray planes and to provide extension information so that producers could examine their fields before damage became severe. The short period during which the methods were used, and the modifications that were instituted from year to year, limit the scope of the evaluation. In addition, information on the distribution of larvae and pupae in fields, on which effective sampling techniques could be developed, is lacking. The following recommendations for survey methods to provide predictions of the location and abundance of bertha armyworm infestations were based on the evaluation presented in this paper:

1. Pupal sampling is expensive in time and effort. Weather conditions may often severely restrict its application. Fall sampling was used to predict the need and thus ensure the availability of insecticides, but information on the abundance of larvae earlier in the summer would probably be sufficient to provide this information. Spring sampling provides a better prediction of the potential population of bertha armyworm than does fall sampling. However, an effective method of estimating the abundance of bertha armyworm adults would be preferable to pupal sampling.

2. The catches of bertha armyworm moths in the small number of light traps used during the 1971 to 1974 outbreak did not provide the basis for accurate predictions of areas of infestation. A much larger number of traps, possibly several per rural municipality, would be needed for such predictions. The time and effort required to sort and

identify the insects caught in the light traps precludes this approach. Traps baited with a sex attractant specific to bertha armyworm have a much greater potential for identifying areas of infestation.

3. Sampling for early larval instars of bertha armyworm was the best method of predicting areas of infestation. Timing is critical for this type of survey because if it is too early, infestations are underestimated and if too late, insufficient time is available to alert the producers in infested areas. The method used in 1974 appears to be basically satisfactory and should be further tested. The areas to be sampled should be selected on the basis of a survey for pupae or trapping of adults. Priority should be given to districts identified as having a high population of bertha armyworm. Within these areas, three to five fields should be sampled per rural municipality.

4. Predictions of bertha armyworm abundance should be made for relatively small areas, e.g. rural municipalities, and these predictions should be based on adequate sampling within these areas. When bertha armyworm populations increase to a level at which sampling should be resumed, the intensity of sampling needed to give satisfactory prediction of the severity of infestation for a rural municipality should be determined.

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INFLUENCE OF NUMBER OF SAMPLES ON THE PRECISION OF LC_{50} DETERMINATIONS: RESPONSE OF THE RED FLOUR BEETLE TO METHYL BROMIDE[1]

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ABSTRACT: The precision of measurement of the response to methyl bromide of the red flour beetle, *Tribolium castaneum* (Herbst), varied with the number of samples used. The precision improved as the number of points used in an experiment was increased. For 9 points, and 20 beetles per point, the precision of the LC_{50} varied from 7.7 to 11.2% of its true value. For 36 points the LC_{50} was determined to within 4.5% of its true value.

INTRODUCTION

When strains of the red flour beetle, *Tribolium castaneum* (Herbst), are screened for tolerance to methyl bromide and other fumigants, it is necessary to obtain a precise estimate of the median lethal concentration (LC_{50}) of each strain for the purpose of comparison (Hoskins and Gordon, 1956). The LC_{50}s should have a predetermined level of precision so that comparisons between LC_{50}s can have validity at the precision level used. In many instances, however, the size of the experiment, and hence the precision of LC_{50}s (or LD_{50}) determination is influenced more by practical considerations, such as the numbers of insects available, than by the experimenter's desire to obtain results with a given level of precision.

Howe (1974) mentions that, in the published literature, the numbers of dosage levels, replicates, and even the numbers of insects used per treated sample (point), have sometimes been too low for the proper determination of the probit lines representing dosage mortality relationships. He suggested that at least seven dosage levels are needed, and that one of these should be chosen to give complete kill with the remainder balanced around the LD_{50}.

Barker (1976a) examined the effect of population densities of 20 and 40 beetles per vial on the susceptibility of the red flour beetle to methyl bromide and found that for the numbers of beetles used per sample, population density did not influence the results of LC_{50} determinations, though precision was better when 40 beetles were used.

The purpose of this paper is to show that the precision of the LC_{50} depends on the number of samples (points) used and that a considerably greater number of points than the 7 suggested by Howe (1974) may be required to give satisfactory levels of precision.

MATERIALS AND METHODS

The strain of red flour beetle used in this experient has been reared at this laboratory for more than 5 years. Adult insects 5 to 13 days old were sieved from a culture medium of unenriched flour and ground brewer's yeast (95:5; w:w). Approximately 20 insects were placed in each of 40 test tubes 8.5 cm long by 1.3 cm in diameter. A 0.5 x 7.0 cm strip of filter paper was placed in each of the 40 test tubes to provide more crawling area for the beetles at the bottoms of the test tubes.

To keep each test tube upright, it was placed in a glass jar which was 8.0 cm long and 5.0 cm in diameter. Each jar with its test tube, was placed in one of 40 sealers of 1.735 litre volume. The 40 sealers were arranged at random into groups of 4. One group of 4 sealers was not treated and the other groups received dosages of methyl bromide which

[1] Contribution No. 842, Agriculture Canada, Research Station, Winnipeg, Manitoba.

ranged from 5.16 to 8.61 mg of methyl bromide per liter. There were 9 dosage levels and each dosage was replicated 4 times, a total of 36 points. The gas was measured into the sealers with a gas burette. The exposure period was 5 hours. This system was similar to the one used by Barker (1976b).

After fumigation, the insects were placed on 8 g of clean rearing medium in vials which were 7 cm long and 3 cm in diameter. A snap-on cap with a hole covered with brass mesh confined the insects to the vials. The vials with the insects were then placed in incubators at 30 ± 0.5°C and about 75% RH for 7 days after which the number of live and dead insects was recorded.

Logit analyses (Ashton, 1972) were performed for each replicate individually and for each combination of replicates. The numbers of points used in the analyses were 9, 18, 27, and 36, depending on the combination of replicates; all possible combinations were examined. Standardized residuals were examined, but no points could be rejected as outliers at 1% per point level. Tests for parallelism of logit lines, and for relative tolerance to methyl bromide were also performed according to Ashton's (1972) methods. The 95% confidence intervals (CI) were determined for each LC_{50}.

The precision of each LC_{50} determination was measured by:

$$A\% = \frac{(LC_{50} + CI) - (LC_{50} - CI)}{LC_{50}} \times 100$$

The numbers of points required to obtain 5% precision were also estimated from a ratio of standard errors using:

$$n = \frac{(A\%)^2}{(5)^2} \times (\text{No. points used for A\%}) = \frac{(\text{SE } LC_{50})^2 \times (\text{No. points})}{(\text{SE } LC_{50} \times \frac{5}{A\%})^2}$$

(M. Bickis, personal communication)[2].

RESULTS AND DISCUSSION

The LC_{50}s, and their corresponding confidence intervals are listed individually for each replicate and for each combination of replicates (Table 1). The LC_{50}s ranged from a low of 6.88 mg methyl bromide per litre in replicate 1, to a high of 7.31 mg/l in replicate 4. At the evenly spaced dosages used, mortalities were also evenly spaced and ranged from about 5% for the lowest dosage to about 95% for the highest dosage level used.

A comparison was then performed on the most widely separated sets of data (replicates 1 and 4) to determine whether or not their regression lines were parallel, and if so, what was the relative tolerance ratio to the fumigant. There was no conflict with the hypothesis of parallelism between the logit lines obtained for replicates 1 and 4 (Table 2), and the tolerance ratio obtained was found to be 1.06 with a standard error of 0.03 (Table 3).

When the number of points included in an analysis was increased from 9 to 18, the LC_{50}s ranged from 6.89 to 7.20 mg/l. An increase to 27 in the number of points included in an analysis, reduced the range covered by the LC_{50}s to between 6.96 and 7.10 mg/l (Table 1).

The width of the confidence interval bands changed with the LC_{50}s. When only 9 points were included in the analysis, replicate 1 had the lowest $LC_{50} - CI$ (6.51 mg/l) whereas replicate 4 had the highest $LC_{50} + CI$ (7.60 mg/l).

An increase to 18 in the number of points used per analysis reduced the span of the confidence intervals to between 6.67 and 7.43 mg/l. The use of 27 points per analysis

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Table 1. The effect of number of samples (points) on the heterogeneity factor and on the precision of the LC_{50} for the red flour beetle, *Tribolium castaneum*, treated with methyl bromide.

Replicates and combinations	LC_{50} mg/l	±95% CI[1]		C.I. Band	Heterogeneity factor	Precision[2]
		9 points				
1	6.88	6.51	7.27	0.76	0.72	11.09
2	6.91	6.59	7.25	0.66	0.48	9.53
3	7.11	6.72	7.52	0.80	1.58	11.23
4	7.31	7.04	7.60	0.56	0.56	7.72
		18 points				
1 + 2	6.89	6.67	7.11	0.44	0.55	6.43
1 + 3	6.99	6.74	7.25	0.51	1.07	7.21
1 + 4	7.07	6.84	7.31	0.47	1.04	6.64
2 + 3	7.00	6.78	7.24	0.46	0.97	6.57
2 + 4	7.09	6.88	7.31	0.43	0.83	6.02
3 + 4	7.20	6.98	7.43	0.45	1.18	6.30
		27 points				
1 + 2 + 3	6.96	6.78	7.15	0.37	0.84	5.34
1 + 2 + 4	7.01	6.84	7.20	0.36	0.83	5.11
1 + 3 + 4	7.08	6.89	7.28	0.39	1.11	5.41
2 + 3 + 4	7.10	6.92	7.28	0.36	1.00	5.06
		36 points				
1 + 2 + 3 + 4	7.04	6.88	7.20	0.32	0.95	4.48

[1] CI = confidence interval.

[2] Precision: LC_{50} determined to within this % of its true value.

Table 2. Chi-square test for parallelism of regression for the response of the red flour beetle, *Tribolium castaneum*, treated with methyl bromide (replicates 1 and 4).

	Sum of squares	df	Mean squares
Parallelism of the regressions	2.79	1	2.79^{NS}
Residual heterogeneity	9.02	14	0.64^{NS}
Total	11.82	15	

^{NS} Not significant.

further reduced the span embraced by confidence interval bands to between 6.78 and 7.28 mg/l. The narrowest confidence interval span was obtained when 36 points were considered.

The width of the confidence interval span influenced the calculation of the precision of each LC_{50} determination. As the confidence interval spans were reduced with increas-

Table 3. The determination of common slope, new intercepts and relative tolerance ratio of the red flour beetle, *Tribolium castaneum,* to methyl bromide (replicates 1 and 4).

Factor	Value
Pooled slope	23.774
Standard error of pooled slope	3.223
New intercept, replicate 1	−19.884
New intercept, replicate 4	−20.564
Tolerance ratio	1.068
Standard error of tolerance ratio	0.030

Table 4. The effect of the number of samples (points) on estimates of the number of points required to obtain standard errors of the LC_{50}'s to within 5% of their true values.

Replicates and combinations	Standard error	Precision[1]	Estimated number of points required
	9 points		
1	0.010181	11.09	44.27
2	0.008749	9.53	32.69
3	0.010309	11.23	45.40
4	0.007089	7.72	21.45
Average			35.95
	18 points		
1 + 2	0.006588	6.43	29.76
1 + 3	0.007393	7.21	37.42
1 + 4	0.006806	6.64	31.74
2 + 3	0.006734	6.57	31.07
2 + 4	0.006162	6.02	26.09
3 + 4	0.006458	6.30	28.57
Average			30.77
	27 points		
1 + 2 + 3	0.005634	5.34	30.79
1 + 2 + 4	0.005390	5.11	28.20
1 + 3 + 4	0.005710	5.41	31.60
2 + 3 + 4	0.005332	5.06	27.65
Average			29.56
	36 points		
1 + 2 + 3 + 4	0.004798	4.48	28.90

[1] **Precision: LC_{50} determined to within this % of its true value.**

ing numbers of points, precision was improved (Table 1) so that 36 points yielded an LC_{50} determination to within 4.48% of its true value. A precision of the LC_{50} within 5% of its true value would have required between 27 and 36 points.

The approximate number of points (n) required to yield an LC_{50} within 5% of its true value was then calculated using the standard errors from each logit analysis. When few points (9) were used, the estimates of n fluctuated from 21 to 45 (Table 4). This range was reduced to between 27 and 31 when 27 points were used. The minimum number of points required to provide the 5% precision was calculated as 29, confirming the suggestion above that between 27 and 36 points were required to obtain an LC_{50} to within this level of precision.

The heterogeneity factors (Table 1) found for all of the replicate combinations were close to 1, indicating that the response of the beetles to the fumigant dosage levels was linear and that the variation between groups was comparable to that expected from a binomial distribution. The heterogeneity factor was not, however, related to precision of the LC_{50} determination.

If the LC_{50}s can be determined to a precision level of 5% or less, small differences between strains or between treatments should easily be detected. Large confidence intervals and poor precision will tend to obscure differences which may be real. Barker (1976b) did not demonstrate differences between the tolerances of successive generations of a strain of flour beetle to methyl bromide even though the LC_{50}s were determined to within about 5% of their true values. If a greater number of points per test had been used, this distinction between generations may have been possible.

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NEW ANNOTATED RECORDS OF ODONATA FROM THE PROVINCE OF MANITOBA WITH NOTES ON THEIR PARASITISM BY LARVAE OF WATER MITES

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ABSTRACT: Twenty-three species of Zygoptera, of which seven, *Lestes forcipatus* Rambur, *L. rectangularis* Say, *Chromagrion conditum* (Hagen), *Enallagma carunculatum* Morse, *E. vernale* Gloyd, *Ischnura posita* (Hagen) and *I. perparva* Selys are reported from Manitoba for the first time, and twenty-seven species of Anisoptera, including two subspecies, of which five, *Aeshna interrupta interrupta* Walker, *A. verticalis* Hagen, *Epitheca canis* MacLachlan, *Sympetrum rubicundulum* (Say) and *S. illotum* (Hagen) are reported from Manitoba for the first time, were recorded. Fifteen species of Zygoptera and eight species of Anisoptera were parasitized by water mite larvae. Larvae for the species *Arrenurus major* Marshall, *A. americanus* Marshall, *A. marshallae* (Piersig), and *Limnochares americana* Lundblad, were the most frequently found as parasites. *L. americana* showed a marked preference for the thorax of the host (97.7%) while the other three species of mites showed no site preference.

INTRODUCTION

As part of a study of the population dynamics of water mites (Acari: Hydrachnellae) of the ponds in the Fort Whyte Nature Reserve, 14.5 km southwest of Winnipeg, collections were made from 1975 to 1977 to establish host-parasite relationships between larvae of water mites and adult Odonata. Supplementary collections of water mites and Odonata were made in selected areas of the Province of Manitoba resulting in this report on new records of Odonata with some comments on their parasitism by larvae of water mites. Some species records for 1978 are included to make this report more complete.

In previous studies, eighteen species of Zygoptera and fifty-five species of Anisoptera have been reported from Manitoba (Walker 1912, 1933, 1941, 1953, 1958; Walker and Corbet 1975). None of these works represent the same degree of intensity of collecting as does the present study.

MATERIALS AND METHODS

The Odonata were collected, using an insect net, 38 cm in diameter, with a 1.5 m handle, from eighteen sites as follows:

1. Fort Whyte Nature Reserve, 14.5 km southwest of Winnipeg.
2. Sturgeon Creek at Saskatchewan Avenue, Winnipeg.
3. Living Pairie Museum, Ness Avenue, Winnipeg.
4. 250 Elm Street, Winnipeg.
5. Wilkes Avenue at Waverly Street, Winnipeg.
6. St. Andrews on the Red, 22 km north of Winnipeg.
7. LaBarriere Park, 16 km south of Winnipeg.
8. Gravel Pits, Bird's Hill.
9. Brokenhead River, at Trans-Canada Highway, Sandilands.
10. Stevenson's Point, Winnipeg Beach.
11. Roadside, 8 km north of the junction of Highways 4 and 10.
12. Jackfish Lake, west of junction of Highways 10 and 45.
13. Clear Lake Campground, Riding Mountain National Park.
14. Audy Lake Road, Riding Mountain National Park.
15. Arrowhead Trail, by Podge Lake, Riding Mountain National Park.

16. Arrowhead Trail, by Beaver Pond, Riding Mountain National Park.
17. Brokenhead River, by Highway 44, near Beausejour (1978 only).
18. Buffalo Compound, Riding Mountain National Park (1978 only).

RESULTS AND DISCUSSION

In the collection made for this study, the Suborder Zygoptera was represented by two families, seven genera and twenty-three species, while the Suborder Anisoptera was represented by four families, ten genera and twenty-seven species (including two subspecies). A total of 1333 Zygoptera (1123 males, 210 females) and 837 Anisoptera (543 males, 294 females) were examined. Seven species of Zygoptera, *Lestes forcipatus* Rambur, L. *rectangularis* Say, *Chromagrion conditum* (Hagen), *Enallagma carunculatum* Morse, *E. vernale* Gloyd, *Ischnura posita* Hagen and *I. perparva* Selys, and five species of Anisoptera, *Aeshna interrupta interrupta* Walker, *A. verticalis* Hagen, *Epitheca canis* MacLachlan, *Sympetrum illotum* (Hagen), and *S. rubicundulum* (Say) were recorded from Manitoba for the first time.

The sequence used in the species list follows that of Walker (1953, 1958) and of Walker and Corbet (1975). In the case of new records for the Province of Manitoba, only records from adjacent areas are cited.

Suborder: Zygoptera
Family: Agriidae

Agrion aequabile (Say)
Site 17: 8 ♂♂, 4 ♀♀, 79.06.24
Previous records from Manitoba: Treesbank; Sandilands; Winnipeg (Walker 1953, p. 73)

Family: Lestidae

Lestes congener Hagen
Site 1: 2 ♂♂, 77.08.08; 1 ♂, 78.08.30.
Site 9: 1 ♂, 77.09.08.
Site 12: 13 ♂♂, 11 ♀♀, 77.08.18.
Previous records from Manitoba: Treesbank; Westbourne (Walker 1953, p. 95).

L. unguiculatus Hagen
Site 1: 1 ♂, 77.08.04; 9 ♂♂, 1 ♀, 77.08.08; 1 ♂, 1 ♀, 77.08.11.
Site 2: 1 ♂, 1 ♀, 77.07.06; 1 ♂, 75.07.14.
Site 9: 2 ♂♂, 77.08.13.
Previous records from Manitoba: Winnipeg; Treesbank; Stony Mountain; Onah; Westbourne (Walker 1953, p. 99).

L. dryas Kirby
Site 10: 1 ♂, 76.06.20.
Previous records from Manitoba: Stockton; Treesbank; Westbourne; Portage la Prairie; Winnipeg; Victoria Beach; Riding Mountain National Park; The Pas (Walker 1953, p. 103).

Notes: This specimen was parasitized by two *Arrenurus major* Marshall larvae and by one *A. americanus* Marshall larva on the thorax behind legs-3.

L. disjunctus disjunctus Selys
Site 1: 1 ♂, 77.06.27; 1 ♂, 77.08.08; 1 ♂, 2 ♀♀, 77.08.11.
Site 2: 1 ♂, 1 ♀, 75.07.23; 1 ♂, 75.08.05.
Site 6: 1 ♂, 77.09.06.
Site 9: 2 ♂♂, 1 ♀, 77.08.13.
Site 12: 18 ♂♂, 3 ♀♀, 77.08.18.
Site 16: 3 ♂♂, 1 ♀, 77.07.31.
Previous records from Manitoba: Treesbank; Westbourne; Winnipeg; Victoria Beach; Riding Mountain National Park; The Pas; Wabowden; Gillam (Walker 1953, p. 107).

Notes: One male *L. disjunctus* from Site 2 (75.08.05) carried a larva of *Arrenurus americanus* on the thorax behind legs-3. *L. disjunctus,* the second most frequently taken species in mid August, peaked at that time. Walker (1953) noted that this species occurred in mid July on the Prairies. The 77.06.27 record from Site 1 is the earliest from either the Prairies or Ontario.

L. forcipatus Rambur
Site 9: 3 ♂♂, 1 ♀, 77.08.13.
Site 16: 1 ♂, 2 ♀♀, 77.07.31.
Previous records from Manitoba: None.

Notes: This species was reported from Ontario (Smoky Falls, Rainy River, Fort Frances) and from Saskatchewan (Prince Albert National Park – Waskesiu Lake) (Walker 1953, p. 112). Mitchell (1959) found *Arrenurus pollictus* Marshall to be a parasite of *L. forcipatus.*

L. rectangularis Say
Site 1: 1 ♂, 77.08.08; 1 ♂, 78.06.26.
Previous records from Manitoba: None.

Notes: This species was reported from Ontario (Minaki, Kenora, Rainy River) (Walker 1953, p. 115). Walker commented that the species "will certainly be found in eastern Manitoba."

Family: Coenagrionidae

Chromagrion conditum (Hagen)
Site 1: 2 ♀♀, 77.06.16.
Previous records from Manitoba: None.

Notes: This species was reported from Ontario (Elgin, Waterloo, Oxford, Peel, York, Simcoe, Carleton, and Renfrew counties; Muskoka, Parry Sound, Nipissing, Algoma, and Cochrane districts; Lake Abitibi is the most northerly record) (Walker 1953, p. 160). A larva of *Limnochares americana* Lundblad was on the thorax behind legs-3 on one of the specimens.

Nehalennia irene (Hagen)
Site 1: 2 ♀♀, 76.07.08.
Site 2: 1 ♂, 75.07.14.
Site 9: 1 ♀, 77.08.13.
Previous records from Manitoba: Treesbank; Onah; Westbourne; Winnipeg; Winnipeg Beach; Victoria Beach; Dauphin; The Pas (Walker 1953, p. 169).

Coenagrion resolutum (Hagen)
Site 2: 14 ♂♂, 1 ♀, 77.06.08; 11 ♂♂, 77.06.15; 1 ♀, 77.06.22.
Site 10: 4 ♂♂, 76.06.20; 3 ♂♂, 76.07.03.
Previous records from Manitoba: Treesbank; Portage la Prairie; Winnipeg; Victoria Beach, Lake Winnipeg; Virden; Dauphin; Lake Winnipegosis; Riding Mountain National Park; The Pas; and northward (along the Hudson Bay Railway) to Churchill (Walker 1953, p. 177).

Notes: Six (18.6%) of the males were parasitized by larvae of *Arrenurus major* and a larva of an unidentified species of the same genus. Most of the larvae (72.7%) occurred on the thorax behind leg-3, while the balance were either on abdominal segments 1 (4.6%) or 6 and 7 (22.7%). *C. resolutum* was the most common damselfly in mid June.

C. angulatum Walker
Site 1: 2 ♂♂, 76.06.05.
Site 10: 1 ♂, 76.06.20.

Previous records from Manitoba: Treesbank; Onah; Portage la Prairie; Virden; Winnipeg Beach and Lake Winnipeg; Victoria Beach; Winnipeg; Dauphin; The Pas (Walker 1953, p. 185).

Notes: One male from Fort Whyte was parasitized by 13 *Arrenurus americanus* larvae (5 on the thorax, 8 on abdominal segment 1).

Enallagma carunculatum Morse

Site 1: 150 ♂♂, 40 ♀♀, 77.06.16 to 77.09.12.

Previous records from Manitoba: None.

Notes: This species was reported from Ontario (Kenora district and eastern Ontario) and Saskatchewan (Echo Lake and Last Mountain Lake, both saline lakes), (Walker 1953, p. 203). Walker commented that the "breaks in distribution in the Prairie Provinces are probably only apparent due to insufficient collecting . . . it will quite certainly be found in eastern Manitoba." 51.3% of the males and 30% of the females were parasitized by *Limnochares americana* (thorax); *Hydrodroma despiciens* (Müller) (abdomen); *Tiphys* n.sp. (thorax between legs-2 and -3); *Arrenurus marshallae* (Piersig), *A. major* and *A. americanus* (thorax and abdomen). Large numbers occurred from July 12 to August 9 with the greatest numbers in the week of July 26. *E. carunculatum* was the most prevalent species from July 19 to August 16. Walker reported the flight period for Ontario as June 19 to September 20 with the maximum numbers in July and early August. For British Columbia, he reported the flight period as June 3 to September 29.

E. civile (Hagen)

Site 1: 185 ♂♂, 18 ♀♀, 77.05.12 to 77.09.13.
Site 2: 5 ♂♂, 2 ♀♀, 77.06.08; 2 ♂♂, 2 ♀♀, 77.06.15; 1 ♂, 75.07.08.
Site 3: 4 ♂♂, 2 ♀♀, 77.09.06.
Site 6: 1 ♂, 77.09.07.
Site 9: 2 ♂♂, 2 ♀♀, 77.08.13.
Site 10: 1 ♂, 76.06.20.

Previous records from Manitoba: Emerson; Winnipeg, Selkirk; Victoria Beach; Lepelletier (Walker 1953, p. 208).

Notes: Eighty-three (41.3%) of the males and six (23%) of the females were parasitized by *L. americana* (thorax), *A. major* and *A. americanus* (thorax and abdomen). This was the second most numerous Odonatan overall and was the most common species in May and the second most common species in June. Two population peaks were noted, one in late May and the second in late June. Walker (1953, p. 208) reported it was the commonest damselfly in Victoria Beach at the end of June, 1931, and that it flew particularly with *E. clausum* Morse. He further noted that the flight period for Ontario was June 6 to September 12. It is of interest to note that the Manitoba flight period is almost a full month longer, but this may have been due to the very warm dry and early spring of 1977.

E. clausum Morse

Site 1: 9 ♂♂, 12 ♀♀, 77.06.05 to 77.08.06.
Site 2: 1 ♀, 77.06.08.

Previous records from Manitoba: Onah; Husavick; Victoria Beach, Lake Winnipeg; Lake Dauphin (Walker 1953, p. 211).

Notes: Five (55.6%) of the males and three (23.1%) of the females were parasitized by *Limnochares americana, Arrenurus major* and *A. americanus* (all on the thorax). The peak numbers occurred in late June. This agrees with the findings of Walker (1953, p. 212).

E. boreale Selys
Site 1: 78 ♂♂, 7 ♀♀, 77.05.30 to 77.09.12.
Site 12: 3 ♂♂, 77.08.18.
Site 16: 1 ♀, 77.07.31.
Previous records from Manitoba: Treesbank; Onah; Stockton; Strathclair; Winnipeg; Winnipeg Beach; Lake Winnipeg; Wabowden; Gillam; Herchmer (Walker 1953, p. 215).

Notes: Thirty-three (40.7%) of the males and two (25%) of the females were parasitized by *Limnochares americana* and *Arrenurus marshallae* (thorax), *A. americanus* and *A. major* (thorax and abdomen). Peak numbers were noted on June 21 and the numbers remained high for the following two weeks. Walker (1953, p. 216) noted that, in Ontario, this species is present for a month to a month and a half ending in early July. He reported the flight period for British Columbia as April 29 to mid-October.

E. cyathigerum (Charpentier)
Site 1: 133 ♂♂, 11 ♀♀, 76.06.02 to 76.08.03.
Site 2: 1 ♂, 77.06.08; 1 ♂, 75.07.14.
Site 12: 36 ♂♂, 3 ♀♀, 77.08.18.
Previous records from Manitoba: Assiniboine River near Treesbank; Onah; Strathclair; Stockton; Winnipeg; Selkirk; Victoria Beach; Lepelletier; Virden; The Pas; Mile 214, Hudson Bay Railway; Wabowden; Gillam (Walker 1953, p. 219).

Notes: Seventy-five (43.9%) of the males and four (28.6%) of the females were parasitized by *Limnochares americana* (thorax), *Arrenurus* sp., *A. major,* and *A. americanus* (thorax and abdomen). Two population peaks of *E. cyathigerum* were noted, the first in the first three weeks of June (it was the most prevalent species on June 7 and June 21) and the second peak on August 23 when it was again the most prevalent species. Walker (1953, p. 220) found it very abundant in early July at The Pas.

E. vernale Gloyd
Site 1: 12 ♂♂, 76.06.07.
Site 3: 1 ♀, 77.09.07.
Previous records from Manitoba: None.

Notes: This species was reported from Ontario (Kenora, Sioux Lookout) and Saskatchewan (Big Quill Lake) (Walker 1953, p. 223). Walker said the flight period "is early and brief." The September record is the latest flight record for this species, but this is not surprising considering the flight records for other species in Manitoba.

E. hageni (Walsh)
Site 1: 183 ♂♂, 16 ♀♀, 77.05.12 to 77.08.12.
Site 2: 39 ♂♂, 4 ♀♀, 76.06.08 to 76.07.23.
Site 10: 7 ♂♂, 76.06.20.
Site 16: 1 ♂, 77.07.31.
Previous records from Manitoba: Westbourne; Winnipeg; Stockton; Treesbank; Onah; Selkirk; Victoria Beach, Lake Winnipeg; Virden; Dauphin; Fork River; Winnipegosis; The Pas (Walker 1953, p. 227).

Notes: One hundred and twenty-two (53.0%) of the males and six (30%) of the females were parasitized by *Limnochares americana* and *Piona* sp. (thorax); *Arrenurus* sp., *A. major,* and *A. americanus* (thorax and abdomen). *E. hageni* was the most prevalent of all the Odonata collected. Large populations were present from June 21 through August 2, with peaks in late June and again in mid July. *E. hageni* was the most prevalent species on June 28 and from July 12 to 19. It was the second most prevalent on June 21 and from July 19 to August 9. Walker (1953) gave the flight period for this species in Ontario as May 30 to August 21.

E. ebrium (Hagen)
Site 1: 33 ♂♂, 5 ♀♀, 77.06.06 to 77.08.08; 1 ♀ (teneral), 77.09.27.
Site 2: 100 ♂♂, 7 ♀♀, 76.06.08 to 76.07.23.
Site 10: 4 ♂♂, 76.06.20; 8 ♂♂, 1 ♀, 76.07.03.
Previous records from Manitoba: Westbourne; Assiniboine River near Treesbank; Onah; Stockton; Victoria Beach, Lake Winnipeg; Selkirk; Winnipeg; Dauphin; Virden; Winnipegosis; Fork River; The Pas (Walker 1953, p. 230).

Notes: Ninety (62.1%) of the males and nine (64.3%) of the females were parasitized by *Limnochares americana, Neumania onondaga* Habeeb, and *Arrenurus auris* Lavers (thorax); by *A. major, A. marshallae, A. americanus,* and *Arrenurus* sp. (thorax and abdomen), and by *A. megalurus intermedius* Marshall (abdomen). Collections of *E. ebrium* were large from June 21 to July 19, with peaks on June 28 and July 12. It was the most prevalent species on July 5. The September 27 record is the latest flight record for Canada. Walker (1953) noted that *E. berium* had two peaks in Toronto – one in mid June and the other in mid July. The present records tend to span these two peak periods. Mitchell (1969) noted *L. americana* on the thorax of *E. ebrium* which he also found to be the "only known host of *A. americanus* (on the thorax)" (Mitchell 1964). Mitchell (1964) also noted that *E. ebrium* was a host for *A. major* (thorax and abdomen). Mitchell (1969) noted that it was not unusual to find two species of *Arrenurus* parasitizing *E. ebrium* at the same time. This was noted for several of the damselfly species during the present study. Five other species of *Arrenurus* (*A. compactilis* Marshall, *A. falcicornis* Marshall, *A. gennadus* Cook, *A. magnicaudatus* Marshall, and *A. americanus mucronatus* Lavers) were reported by Mitchell (1964) on *E. ebrium.*

Ischnura posita (Hagen)
Site 1: 1 ♀, 76.06.17.
Previous records from Manitoba: None.

Notes: This species was reported from Southern Ontario, Michigan and Minnesota (Walker 1953, p. 256). Both Munchberg (1951) Mitchell (1964) reported *Arrenurus major* as a parasite of both thorax and abdomen.

I. verticalis (Say)
Site 1: 7 ♂♂, 2 ♀♀, 76.06.05 to 76.07.28.
Previous records from Manitoba: Winnipeg; Victoria Beach (Walker 1953, p. 260).

Notes: Five (71.4%) of the males were parasitized by *Limnochares americana* and *Arrenurus major* (thorax), and by *A.* sp. (abdomen). Mitchell (1959, 1964) reported *A. major* parasitizing 90 to 95% of *I. verticalis* adults with a mean of 13 larvae on each. Munchberg (1953) stated that *A.* sp. parasizized *I. verticalis* on the thorax, and occasionally near the middle of the abdomen.

I. perparva Selys
Site 1: 1 ♂, 76.06.24; 1 ♀, 76.07.12; 28 ♀♀, 78.05.25 to 78.06.19.
Site 10: 2 ♂♂, 76.06.20; 1 ♂, 76.07.03.
Previous records from Manitoba: None.

Notes: This species was reported from British Columbia (Walker 1953, p. 263). Whitehouse (1941) noted that it is "seldom found far from the weedy banks of a pond or stream." Three (75%) of the males were parasitized by *Limnochares americana* and *Arrenurus major* (thorax).

I. damula Calvert
Site 1: 2 ♀♀, 76.06.24; 1 ♂, 1 ♀, 76.06.08; 1 ♀, 76.07.12; 1 ♀, 77.08.11; 1 ♀, 76.08.12.
Previous records from Manitoba: Kildonan Park, Winnipeg; Whiteshell Provincial Park, 12 miles west of the Ontario border on the Trans-Canada Highway; Victoria Beach (Walker 1953, p. 270).

Notes: Three (50%) of the females were parasitized by *Limnochares americana* (thorax); by *Arrenurus americanus* and *A. major* (abdomen). Walker (1953, p. 271) found the flight period for *I. damula* to be June 28 to August 7. The present study extends these dates by a week.

Suborder: Anisoptera
Family: Aeshnidae

Aeshna eremita Scudder
Site 16: 1 ♂, 77.07.31.
Previous records from Manitoba: Husavick; Treesbank; Onah; The Pas; Lake Atikameg; Blue Lakes (Duck Mountains); Gillam; Mile 214, Hudson Bay Railway (Walker 1958, p. 59).

A. interrupta interrupta Walker
Site 4: 1 ♂, 76.07.25.
Site 14: 2 ♂♂, 76.08.19.
Previous records from Manitoba: None.

Notes: This species was previously recorded from Ontario (Kenora district) and from Michigan (Walker 1958, p. 64).

A. i. lineata Walker
Site 1: 6 ♂♂, 77.08.08; 2 ♂♂, 77.08.11.
Site 4: 1 ♀, 76.07.01; 1 ♂, 76.07.05; 1 ♀, 76.07.23; 1 ♂, 4 ♀♀, 76.07.24; 7♂♂, 5 ♀♀, 76.07.25; 2 ♀♀, 76.07.27; 1 ♂, 76.07.28.
Site 8: 4 ♂♂, 4 ♀♀, 76.07.10.
Site 12: 3 ♂♂, 1 ♀, 77.08.18.
Site 16: 1 ♂, 1 ♀, 77.07.31.
Previous records from Manitoba: Treesbank; Winnipeg; Winnipeg Beach and Victoria Beach, Lake Winnipeg; Husavick; Westbourne; Stockton; Riding Mountain National Park; Swan River; The Pas; Lake Atikameg; Pitwitonei; Blue Lakes (Duck Mountains) (Walker 1958, p. 66).

Notes: The maximum numbers were noted in late July and early August. Large numbers were noted at Site 4 during 1976 but none were seen in 1977. Walker (1958, p. 67) found the species most abundant in July and August.

A. canadensis Walker
Site 4: 1 ♀, 76.07.20.
Site 13: 1 ♂, 77.08.01.
Previous records from Manitoba: Westbourne; Treesbank; Victoria Beach; Winnipeg; Husavick; Township of Ronge; Lake Atikameg (Walker 1958, p. 72).

A. verticalis Hagen
Site 4: 1 ♀, 76.07.20.
Previous records from Manitoba: None.

Notes: This species was reported from Ontario (Essex, York, Bruce, Simcoe, Leeds, and Carleton counties; Muskoka district) and from Minnesota (Walker 1958, p. 76).

Anax junius (Drury)
Site 1: 22 ♂♂, 6 ♀♀, 77.05.12 to 77.07.04; very many others seen flying, either singly or in pairs, during this period.
Site 2: 1 ♂, 77.06.08; 1 ♂, 1 ♀, 77.06.15; 1 ♂, 77.06.22; 3 ♂♂, 77.07.06.
Site 9: 2 ♀♀, 77.08.13.
Previous records from Manitoba: Treesbank (Aweme); Stockton; Kildonan Park, Winnipeg; Victoria Beach, Lake Winnipeg; Husavick (Walker 1958, p. 127).

Notes: One male (Site 1, 77.06.20) was parasitized on the thorax by a larva of *Hydrodroma despiciens* and three of *Arrenurus major.* The combination of a warm

spring and strong southerly winds may have helped this migratory dragonfly to arrive so early and in such large numbers in Manitoba in 1977. The species was not seen either in 1975 or 1976. Whitehouse (1941) noted that the "species was far from common in the west."

Family: Gomphidae

Gomphus (Gomphorus) graslinellus Walsh
Site 1: 20 ♂♂, 11 ♀♀, 77.05.26 to 77.07.07.
Previous records from Manitoba: Aweme (treesbank) (Walker 1958, p. 213).

Notes: An early summer species characterized by a peculiar "switchback" flight. Our record for 1977.05.26 is the earliest for Canada. One female was found emerging on 1977.06.02, and copulating pairs were taken from June 20 to July 7. The peak flight period was late June.

G. (G.) cornutus Tough
Site 1: 1 ♂, 1 ♀, 77.06.23; 1 ♂, 77.06.27.
Previous record from Manitoba: Red River, Kildonan Park, Winnipeg (Walker 1958, p. 241).

Notes: One male, taken on 77.06.27, was parasitized between legs-2 by a larva of *Limnochares americana.*

Gomphus (Gomphorus) externus Hagen
Site 1: 1 ♂, 77.06.27.
Previous records from Manitoba: Aweme (Treesbank); Winnipeg; Winnipeg Beach, Lake Winnipeg (Walker 1958, p. 250).

Family: Corduliidae

Epitheca spinigera (Selys)
Site 1: 7 ♂♂, 76.06.10; 36 ♂♂, 3 ♀♀, 78.05.29 to 78.06.26.
Site 4: 1 ♂, 78.06.04.
Site 18: 3 ♂♂, 76.06.18.
Previous records from Manitoba: Stockton; Treesbank; Aweme; Onah; Winnipeg; Victoria Beach; Lake Winnipeg; The Pas (Walker and Corbet 1975, p. 52).

Notes: Three males (76.06.10) were parasitized: one by 52 larvae of *Arrenurus marchallae* on abdominal segments 6-9; a second by 46 larvae of *A. marshallae* on abdominal segments 2, 3, 7, 8, 9; and one by one larva of *Arrenurus* sp. on abdominal segment 7.

Epitheca canis MacLachlan
Site 18: 1 ♂, 78.06.88.
Previous records from Manitoba: None.

Notes: Walker and Corbet (1975, p. 55) reported this species from Stanley Rapids, Churchill River, Saskatchewan, and from the Kenora District, Ontario.

Somatochlora ensigera Martin
Site 17: 1 ♂, 78.06.24.
Previous records from Manitoba: Onah; Westbourne (Walker and Corbet 1975, p. 89).

Cordulia shurtleffi Scudder
Site 13: 2 ♀♀, many flying, 78.06.17.
Previous records from Manitoba: Treesbank; Aweme; Onah; Douglas Lake; The Pas; Mile 17, Hudson Bay Railway; Gillam; Churchill (Walker and Corbet 1975, p. 136).

Family: Libellulidae

Libellula quadrimaculata Linne
Site 1: 38 ♂♂, 12 ♀♀, 78.05.23 to 78.06.26.

Site 2: 4 ♂♂, 5 ♀♀, 77.06.08; 1 ♂, 77.06.22.
Site 10: 2 ♂♂, 76.06.20.
Site 13: 1 ♂, 78.06.17.
Site 18: 1 ♂, 78.06.18.

Previous records from Manitoba: Treesbank; Aweme; Onah; Winnipeg; Stony Mountain; Victoria Beach; The Pas (Walker and Corbet 1975, p. 167).

Notes: Walker and Corbet reported the peak periods for Quebec and Ontario as late June and early July.

L. pulchella Drury
Site 1: 1 ♂, 77.06.24; 1 ♂, 77.06.27; 1 ♂, 77.07.04; 8 ♂♂, 78.06.22; 12 ♂♂, 78.06.26.
Site 2: 1 ♂, 1 ♀, 77.07.06.

Previous records from Manitoba: Treesbank; Aweme; Stockton; Onah; Selkirk; Husavick; Winnipeg Beach (Walker and Corbet 1975, p. 182).

Notes: This dragonfly appears to have a very short flight period in Manitoba.

Sympetrum corruptum (Hagen)
Site 1: 41 ♂♂, 14 ♀♀, 77.05.25 to 77.07.21.
Site 2: 1 ♂, 77.06.08; 1 ♂, 1 ♀, 77.06.15.
Site 3: 1 ♂, 77.09.07
Site 12: 1 ♂, 2 ♀♀, 77.08.18.

Previous records from Manitoba: Treesbank; Aweme; Stockton; Onah; Winnipeg; The Pas; Victoria Beach (Walker and Corbet 1975, p. 206-207).

Notes: One male (Site 1 – 77.06.06) was parasitized by a larva of *Hydrodroma despiciens* on the thorax behind legs-3. The peak period was at the end of June with further isolated occurrences from mid August to September. These results agree with those of Walker and Corbet (pp. 207-208) who notes two peaks, the first in late June and early July, and the second in late August and early September. The flight period for Manitoba (Aweme) was June 7 to September 8 (Walker 1933).

S. illotum (Hagen)
Site 1: 1 ♂, 77.05.23.

Previous records from Manitoba: None.

Notes: This species is characterized by seven antenodal cross-veins in the fore-wing, and five in the hind-wing (Walker and Corbet 1975, p. 202). It is known previously from British Columbia (Walker and Corbet p. 210) where it has been found mainly on Vancouver Island with only one record from the mainland. All other records are from the western United States.

S. costiferum (Hagen)
Site 1: 20 ♂♂, 25 ♀♀, 77.07.07 to 77.09.20.
Site 2: 2 ♂♂, 1 ♀, 76.07.06.
Site 4: 1 ♂, 77.08.28.
Site 5: 1 ♀, 77.09.10.
Site 6: 2 ♂♂, 77.09.06.
Site 9: 4 ♂♂, 10 ♀♀, 77.07.06.
Site 11: 2 ♂♂, 1 ♀, 77.08.18.
Site 12: 29 ♂♂, 21 ♀♀, 77.08.18.
Site 13: 3 ♂♂, 1 ♀, 77.07.31.
Site 14: 1 ♀, 76.08.19.

Previous records from Manitoba: Treesbank; Aweme; Stockton; Onah Carberry; Winnipeg; Westbourne; Riding Mountain National Park; Blue Lakes (Duck Mountains) (Walker and Corbet 1975, p. 217).

Notes: This species peaks in numbers in mid-August to September. Walker and Corbet (1975, p. 217) noted that it was one of the latest species to appear and gave the

latest flight dates (outside British Columbia) as September 16 (Nipissing, Ontario). They also noted that adults were "most numerous in August".

S. danae Sulzer, 1776

Site 1: 2 ♂♂, 77.09.12; 3 ♀♀, 77.09.13; many flying, 77.09.20; 3 ♂♂ 2 ♀♀ 77.09.27.
Site 7: 1 ♀, 77.09.07.

Previous records from Manitoba: Treesbank; Aweme; Stockton; Westbourne; Winnipeg; Neepawa; Grandview; The Pas; Wabowden; Blue Lakes (Duck Mountains) (Walker and Corbet 1975, p. 225).

Notes: This species was found mostly in September. This agrees with the findings of Robert (1963) who found that in Quebec, adults rarely appeared before the end of August. Corbet (1962) suggested that *S. danae* may hibernate as an adult. The July 6 records from Sites 2 and 9 are the earliest from Manitoba.

S. internum Montgomery

Site 1: 29 ♂♂, 12 ♀♀, 77.06.23 to 77.08.11.
Site 2: 1 ♀, 77.06.08; 1 ♂, 1 ♀, 77.06.15; 10 ♂♂, 1 ♀, 77.07.06.
Site 3: 1 ♂, 77.09.07.
Site 4: 2 ♀♀, 76.07.21; 5 ♂♂, 76.07.29; 7 ♂♂, 76.08.26; 1 ♂, 3 ♀♀, 76.08.28; 2 ♂♂, 76.09.08.
Site 9: 22 ♂♂, 15 ♀♀, 77.08.13.
Site 11: 2 ♂♂, 1 ♀, 77.08.18.
Site 16: 6 ♂♂, 5 ♀♀, 77.07.31.

Previous records from Manitoba: Deloraine; Stockton; Treesbank; Aweme; Onah; Grandview; Winnipeg; Portage la Prairie; Westbourne; Teulon; Lake Dauphin; The Pas; Wabowden; Gillam (Walker and Corbet 1975, p. 231).

Notes: One female from Site 2 (77.06.08) was parasitized on abdominal segment 4 by two deutonymphs of *Rhizoglyphys spinitarsa* (Hermann) and on thoracic segment 2 by 18 larvae of *Arrenurus americanus.* A male from Site 2 (77.07.06) was parasitized on thoracic segment 3 by two larvae of *A. americanus.* Two peaks of abundance of *S. internum* were noted, the first in late June and early July and the second in mid to late August. Walker and Corbet (p. 231) reported the flight period as June 21 to September 18 at Grandview, Manitoba.

S. obtrusum (Hagen)

Site 1: 2 ♂♂, 1 ♀, 77.07.21; 1 ♀, 77.07.25; 9 ♂♂, 2 ♀♀, 77.08.08; 16 ♂♂, 2 ♀♀, 77.08.11; 1 ♂, 1 ♀, 77.09.12.
Site 4: 1 ♀, 76.07.24; 2 ♂♂, 2 ♀♀, 76.07.28; 1 ♂, 1 ♀, 76.08.28; 1 ♀, 76.09.06.
Site 9: 44 ♂♂, 26 ♀♀, 77.08.13.
Site 12: 3 ♂♂, 77.08.18.
Site 14: 1 ♀, 77.08.19.
Site 15: 1 ♀, 75.07.19.
Site 16: 2 ♂♂, 77.07.31.

Previous records from Manitoba: Westbourne; Swan River (Walker and Corbet 1975, p. 233).

Notes: *S. obtrusum* peaked in numbers in mid August. Walker and Corbet (1975, p. 234) noted that it peaked in late July in Ontario. One female from Site 1 (77.07.25) was parasitized on abdominal segment 6 by two larvae of *Arrenurus major.*

S. rubicundulum (Say)

Site 4: 1 ♂, 76.07.29.
Site 16: 3 ♂♂, 2 ♀♀, 77.07.31.

Previous records from Manitoba: None.

Notes: This species was reported from Michigan and Southern Ontario (Nipissing and Cochrane districts) (Walker and Corbet 1975, p. 229).

Leucorrhinia borealis Hagen
Site 1: 2 ♂♂, 78.05.23; 1 ♀, 78.05.25; 1 ♂, 78.06.26.
Site 13: 1 ♂, 6 ♀♀, 78.06.17.
Site 14: 2 ♂♂, 78.06.18.
Previous records from Manitoba: Ninette; Pelican Lake; Stockton; Aweme; Bird's Hill; Lake Dauphin; The Pas (Walker and Corbet 1975, p. 243).

L. hudsonica (Selys)
Site 4: 1 ♂, 76.07.25.
Previous records from Manitoba: Sandilands; Winnipeg; Portage la Prairie; Winnipeg Beach; Victoria Beach; Dauphin Lake; The Pas; Norway House; Wabowden; Gillam; Herchmer; Churchill; Miles 200, 332 and 412, Hudson Bay Railway (Walker and Corbet 1975, p. 245).

Notes: This specimen was parasitized by 31 larvae of *Arrenurus americanus* all of which were on the thorax behind legs-3.

L. glacialis Hagen
Site 1: 1 ♂, 78.06.22.
Previous records from Manitoba: Aweme (Walker and Corbet 1975, p. 250).

L. proxima Calvert
Site 1: 2 ♂♂, 78.06.22; 1 ♀, 78.06.26.
Site 13: 2 ♀♀, 78.06.18.
Previous records from Manitoba: Aweme; Onah; Winnipeg Beach; The Pas; Gillam (Walker and Corbet 1975, p. 253).

L. intacta (Hagen)
Site 1: 44 ♂♂, 49 ♀♀, 78.05.23 to 78.06.26.
Site 2: 1 ♂, 77.06.08; 1 ♂, 77.06.15; 1 ♂, 77.07.06.
Site 10: 12 ♂♂, 76.06.20.
Previous records from Manitoba: Ninette; Stockton; Aweme; Winnipeg Beach; Victoria Beach (Walker and Corbet 1975, p. 259).

Notes: One male from Site 10 (76.06.20) was parasitized by one *Limnochares americana* larva on the thorax behind legs-3. Mitchell (1964, 1967) found *A. reflexus* Marshall parasitized about 33% of *L. intacta,* all on the abdomen.

Pantala flavescens (Fabricius)
Site 2: 1 ♀, 76.06.08.
Previous records from Manitoba: Husavick (Walker and Corbet 1975, p. 277).

Notes: Most of the records for this species in Canada are for August and September with the flight range of July 13 to September 12 (Walker and Corbet 1975, p. 277). The present record is the earliest flight record for Canada.

These records raise to 82 (25 Zygoptera, 57 Anisoptera) the total number of species of Odonata known from Manitoba. The 33 species and two subspecies of Odonata recorded from Manitoba by Walker (1953, 1958) and Walker and Corbet (1975) but *not* recorded during the present study are listed in Table 1. Many of these species are known only from the far north of Manitoba in areas not visited by us.

The distribution, on the host, of the larvae of the eleven species of water mites recorded as parasites of Odonata in Manitoba are summarized in Table 2. *L. americana* showed a marked preference for the thorax. None of the other frequently-found mites showed a similar or other preference.

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Table 1. Species of Odonata previously reported from Manitoba, but not recorded during the present survey.

Agrion maculatum Beauvois	*Somatochlora walshii* (Scudder)
Coenagrion interrogatum (Hagen)	*S. minor* Calvert
Aeshna juncea Linne	*S. williamsoni* Walker
A. subarctica Walker	*S. fanklini* (Selys)
A. sitchensis Hagen	*S. kennedyi* Walker
A. septentrionalis Burmeister	*S. forcipata* (Scudder)
A. umbrosa umbrosa Walker	*S. whitehousi* Walker
A. constricta Say	*S. septentrionalis* (Hagen)
Hagenius brevistylus Selys	*S. albicincta* (Burmeister)
Ophiogomphus colubrinus Selys	*S. hudsonica* (Selys)
O. rupinsulensis (Walsh)	*S. cingulata* (Selys)
Gomphus exilis Selys	*Libellula julia* Uhler
G. fraternus fraternus (Say)	*Pachydiplax longipennis* (Burmeister)
G. fraternus manitobanus Walker	*Leucorrhinia patricia* Walker
G. notatus Rambur	*L. frigida* Hagen
Macromia illinoiensis Walsh	*Pantala hymenea* (Say)
Williamsonia fletcheri Williamson	*Sympetrum madidum* (Hagen)

(After Walker (1953, 1958), Walker and Corbet (1975)).

ADDENDA

Since this paper was prepared five other species of Anisoptera were reported to us by various collectors. All were previously taken in Manitoba. This raises the number of species in the present paper to 62 Anisoptera.

Aeshna constricta Say

Site 1: 1 ♂, 78.07.17; 1 ♂, 1 ♀, 78.08.28.
Site 7: 1 ♂, 1 ♀, 78.08.18.
Gull Lake: 1 ♂, 78.09.04.
Previous records from Manitoba: Westbourne; Treesbank; Magnus; Winnipeg Beach (Walker 1958, p. 111).

Notes: All were caught by J. C. Conroy and J. L. Kuhn.

Hagenius brevistylus Selys

Starr Lake: nymphal exuva, 69.07.20 (collected by D. H. Thorpe).
Black River at Highway 304: 1 ♂, 78.07.29 (collected by W. B. McKillop).
Previous records from Manitoba: Berens River (Walker 1958, p. 147).

Gomphus fraternus fraternus (Say)

Dorothy Lake: 4 ♂♂, 5 ♀♀, 78.07.02 (collected by J. C. Conroy).
Previous records from Manitoba: Victoria Beach, Lake Winnipeg; Berens River (Walker 1958, p. 245).

Macromia illinoiensis Walsh

Starr Lake: 1 nymphal exuva, 78.09.23 (collected by R. Berg).
Previous records from Manitoba: Berens River (Walker and Corbet 1975, p. 26); Walker and Corbet (1975, p. 26) also reported *M. illinoiensis* from the Kenora district.

Somatochlora williamsoni Walker

Gull Lake: 1 ♂, 78.08.04 (collected by J. C. Conroy).
Previous records from Manitoba: Winnipeg Beach (Walker and Corbet 1975, p. 86).

Table 2. Number and location on host of larvae of various species of water mites parasitic on Odonata in Manitoba.

Species of Mites	T-1*	T-2	T-3	A-1	A-2	A-3	A-4	A-5	A-6	A-7	A-8	A-9	A-10	Total	Number of hosts
Hydrodroma despiciens (Müller)	–	1	–	–	–	–	–	–	–	–	–	–	–	1	1
Limnochares americana Lundblad	69	628	76	–	–	–	–	7	9	2	–	–	–	791	281
Neumania onodaga Habeeb	1	–	–	–	–	–	–	–	–	–	–	–	–	1	1
Tiphys haliki Conroy	–	1	–	–	–	–	–	–	–	–	–	–	–	1	1
Piona sp.	–	6	5	–	–	–	–	–	–	–	–	–	–	11	3
Arrenurus sp.	–	27	9	–	–	–	–	–	40	48	2	–	–	126	32
A. auris Lavers	–	5	1	–	–	–	–	–	–	–	–	–	–	6	3
A. americanus Marshall	9	100	37	8	1	–	2	18	96	133	35	6	1	446	65
A. major Marshall	34	457	381	1	1	4	25	203	450	430	45	2	–	2033	184
A. marshallae (Piersig)	–	40	2	–	–	–	6	11	35	21	1	–	–	116	12
A. megalurus intermedius Marshall	–	–	–	–	–	–	2	4	4	12	–	–	–	22	1
Totals:	113	1265	511	9	2	4	35	243	634	646	83	8	1	3554	530

*T-1, T-2, T-3, refer to thoracic segments 1, 2, 3; similarly, A-1, A-2 etc., refer to abdominal segments 1, 2, 3, etc.

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EXOTIC ANTS IN WINNIPEG[1]

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ABSTRACT: Nine species of tropical or sub-tropical ants, *Monomorium pharaonis* (Linnaeus), *Paratrechina fulva* (Myr.), *Pheidole anastasii* Emery, *Solenopsis geminata* (Fabricius), *Solenopsis texana* Emery, *Strumigenys rogeri* Emery, *Tetramorium guineese* (Fabricius), *T. simillimum* (F. Smith), and *Wasmannia auropunctata* (Roger), were found in the Tropical House, Assiniboine Park, and one, *Tapinoma melanocephalum* (Fabricius), in an apartment building in Winnipeg, Manitoba. Brief comments on each species are given. These species are unlikely to become serious pests in Winnipeg as none are cold hardy and most cannot tolerate the low humidity of heated buildings in winter.

INTRODUCTION

The Tropical House, Assiniboine Park, Winnipeg, Manitoba was built to provide the public with an example of a natural tropical environment. An attempt was made to develop a natural ecosystem and in addition to the many plants, animals such as fish, reptiles, birds and mammals form a part of the scene. Inadvertently, the setting has become more natural than originally planned in that many species of insects, including some pests, have apparently been brought in with the plants. Control of the insects with fumigants or insecticide sprays is not possible because of the presence of the other animals. Consequently, some pest species have become well established. Among the ants, one species has increased to the point where maintenance workers have complained of being stung. Following are brief comments on nine species of exotic ants found in the Tropical House and on one species taken from an apartment block in Winnipeg.

MATERIALS AND METHODS

Wherever possible, ants for study were obtained in nest series. In the Tropical House, foraging ants were followed to their nests. For soil-nesting species, soil samples were taken with a small trowel, placed in plastic bags, and later sorted in the laboratory; for plant-nesting species, ants were collected individually with forceps at the nest openings and placed directly in 70% alcohol. The species found in the apartment building were reported to the author by a pest control operator; a number of stray foraging worker ants were collected and preserved in alcohol after the main colony was destroyed.

RESULTS

Monomorium pharaonis (Linnaeus). Pharaoh's ant. This ant is one of the most common and best known of all house-infesting ants and probably occurs in every town or city of commercial importance in the United States (Smith 1965). It has been in Winnipeg at least 35 years as evidenced by preserved speciments dated 1943 in the insect collection of the University of Manitoba. This species is thought to be native of India but has been distributed so widely by commerce that its origin is now uncertain. In North America, it is found under field conditions in southern Florida only.

These ants are omnivorous, but show a slight preference for grease, fats and meat (Smith 1965). Consequently, very large populations can develop in areas where food is handled or prepared, such as in kitchens of restaurants and cafeterias or in bakeries and other similar places. In Winnipeg, though this species has been present for many years, it

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has not become a major pest. It requires a relative humidity of 90% or more for nesting (Peacock *et al.* 1950); it is unlikely that these conditions are common in buildings heated in winter in Winnipeg. In the Tropical House, ants of this species are particularly numerous in the work areas where food is prepared for the animals, but they are also found in lesser numbers throughout the greenhouse area.

Paratrechina fulva (Mayr.). This species has sometimes been referred to as the sugar ant. Creighton (1950) lists nine species of *Paratrechina* in the United States of which five, including *P. fulva* from South America, are introduced. These species are established outdoors in the extreme southern United States, but most are found sporadically in greenhouses throughout the country.

In the Tropical House, this species was found under rocks or stepping stones in the greenhouse area and although no complete nests or queens were found the presence of winged females would indicate that it is established. Smith (1965) states that in this genus the newly fertilized females are capable of establishing new colonies unassisted. These ants undoubtedly eat some of the food supplied for other animals, but since the worker ants neither bite nor sting and as they do no harm to plants, animals or structural materials they can only be considered a nuisance, at most, in the Tropical House.

Pheidole anastasii Emery. Originally described from Costa Rica this species was recorded as established in Florida, in 1933, but had previously been seen in greenhouses in Washington D.C., New Jersey and Illinois (Smith 1933). Obviously it has been present in North America for some time and it appears easily transported by commerce. However, this species is not mentioned in any of the references on pest ants and is unlikely to be a serious problem. It was not abundant in the Tropical House. Only one collection of workers was obtained from the soil in the greenhouse and no evidence of queens or brood could be found.

Solenopsis geminata (Fabricius). Fire ants. This species is native to the southern United States although its main range lies in Central America and the Antilles (Creighton 1950). It is distributed throughout Florida and in an area up to 150 miles from the coast extending from Texas to South Carolina. Until the introduction of the imported fire ants, *S. richteri* Forel in the 1920's and *S. invicta* Buren in the 1930's, *S. geminata* was the most common and noxious of the fire ants in Florida (Buren 1972).

The degree to which *S. geminata* is established in the Tropical House is questionable. Normally this species is extremely polymorphic with the head of the major workers being disproportionately large. In these collections only minor workers were found in the soil suggesting that either the colonies were young or conditions were adverse and prevented the full development of the colonies.

Solenopsis texana Emery. The main distribution of *S. texana* is in the southeastern United States, but it is found under natural conditions as far north as southern Ontario (Creighton 1950). There are no reports of this species as a pest in buildings. Only one specimen of this species was found in the soil of the Tropical House and, therefore, it is not certain whether it is established as a breeding colony or if a few stray workers were brought in with plant material.

Strumigenys rogeri Emery. This species was originally described from West Indian material but is believed to be of West African origin. Creighton (1950) does not list this species, even as an introduction, in the ant fauna of North America, and Brown (1962) cites greenhouses in England and Scotland as the only temperate zone records. Since practically all the plants in the Tropical House have been imported from Florida it appears that *S. rogeri*, if not fully established, is at least present in that state.

The status of this species in the Tropical House is not known. Only two specimens were obtained and these were in soil with other collections.

Tapinoma melanocephalum (Fabricius). This species, which has been introduced to Florida, is thought to be of African or Oriental origin (Smith 1965). It is not a noted pest, but is occasionally found in greenhouses and other heated buildings in the northern

parts of the United States (Smith 1965). In Winnipeg, the only record of this species is from an apartment block situated along the Assiniboine River. The nest was in a steam iron. When the iron was preheated, hundreds of the ants carrying "eggs" (probably larvae and pupae) streamed out the steam holes in the bottom of the iron. Measures were taken to control the ants, but for several weeks afterwards isolated worker ants were found in the kitchen sink, presumably in search of water. However, as the ants were reported to have disappeared it is assumed the main colony was destroyed.

The origin of this colony remains a mystery. According to the tenant no one from the apartment in which the ants were found had been on any trips where these ants might have been picked up and no items such as tropical plants or other material from the tropics had been brought in. The colony appeared mature and since the steam iron was used regularly, the ant colony obviously moved there after developing elsewhere.

Tetramorium quineese (Fabricius) and *T. simillimum* (F. Smith). Both species are noted as "tramp" species and are now so widely distributed that their exact area of origin is questionable. It is generally thought that both are of African origin (Creighton 1950).

T. quineese is more common than *T. simillimum* in North America and is established in the southern United Stated. It ranges from southern Florida to Texas and is particularly common in the Gulf Coast region (Smith 1965). This area of naturalization is a source of infestation for greenhouse populations which have been reported from many parts of the United States. *T. simillimum* appears limited to the most southern parts of Florida. Creighton (1950) questions if it truly has become established as it requires some degree of winter protection even in that part of the state.

Both these species are well established in the greenhouse area of the Tropical House. Numerous colonies were found nesting under stones and in the soil and most had brood and/or many winged sexuals in the nest.

Wasmannia auropunctata (Roger). The little fire ant. This species is neotropical in origin and is extremely common throughout Central and northern South America, the West Indies and the warmer parts of Mexico (Wheeler 1929). Unlike most species of ants, this species does not build a definitive nest: slight shelter and the presence of moisture seem to be the only requirements for nesting. Colonies are often established in spaces between plants and soil, under leaf stalks of palms and other plants with petiole leaf sheaths, or in any natural cavity or confined space in plants (Spencer 1941). For this reason, they are readily dispersed with commercial shipments of tropical plants. As early as 1907, *W. auropunctata* was reported as being one of the most abundant of all species of ants in the greenhouses at Kew Gardens, England (Wheeler 1929). The first outdoor infestation of this species in Florida was found in 1924 (Wheeler 1929) and by 1935 it had become a pest in citrus groves in the southern part of the state (Spencer 1941). However, the species appears strictly tropical and its natural distribution in North America has not extended beyond southern Florida.

The common name, little fire ant, is taxonomically misleading as all other "fire ants" belong to the genus *Solenopsis* to which the genus *Wasmannia* is only distantly related (Smith 1965). The sting of the little fire ant, however, is very similar to that of the true fire ants. Spencer (1941) reports that for many people the sting is very painful at first and later itches intensely for a period of up to 3 days. Numerous stings (12 or more) over a short period may cause the victim to become pale and shaky. For this reason, premium wages are often paid to workers in fruit groves where infestations of this ant are present. However, unlike ants of the genus *Solenopsis,* those of *Wasmannia* do not usually sting unless pressed against the skin by constrictions in clothing such as collars or belts.

W. auropunctata was the most numerous of the ant species in the greenhouse area of the Tropical House. The workers foraged on most of the plants and foraging trails indicated that many colonies were probably present in the petiole sheaths of palms. Colonies found under stones and in the soil had brood and many winged sexuals.

DISCUSSION

This investigation was made approximately 2½ years after the Tropical House was established and it is unlikely that a balance in the ant population had been reached at that time. On the basis of these observations, however, it would seem that the two most troublesome species, *M. pharaonis* and *W. auropunctata,* will be a part of the final species complex. *M. pharaonis,* which can be a serious problem in establishments processing food for human consumption, is likely to be only a nuisance in these surroundings. It does not damage plant or structural materials and its infestation of the animal food does not render the food unusable. Because of its sting, *W. auropunctata* is probably a more serious problem, but again, this species causes no plant or structural damage and is not likely to cause any upsets in the ecological system as designed for public display. The public, being limited to walkways in the Tropical House, is unlikely to encounter this ant and therefore the problem is basically internal, affecting the maintenance personnel only. Unless a person shows a severe allergic reaction to the sting of this ant, as with any native insect, it is merely an annoyance.

The possibility that the Tropical House could be a source of infestation for other buildings in Winnipeg is considered minimal. *M. pharaonis,* the most adaptable of the species found, has not become a serious pest. The species discussed are unlikely to survive in buildings heated in winter because of the low humidity levels that generally prevail. Commercial greenhouses, which could provide suitable environmental conditions for many of these ants, are generally fumigated on a regular basis thereby preventing the development of permanent populations.

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THE EFFECT OF QUEEN-RELATED PROBLEMS AND SWARMING ON BROOD AND HONEY PRODUCTION OF HONEY BEE COLONIES IN MANITOBA[1]

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ABSTRACT: Colonies with queen loss, queen failure or those which swarmed, produced less honey than did normal colonies. There was also less brood reared in colonies in which there was queen loss or queen failure, but brood production, up to the time of swarming, was 16% greater in colonies that swarmed than in normal colonies. On average, honey production in colonies with queen-related or swarming problems was only 58.5% of normal, showing the importance of eliminating these problems whenever possible.

INTRODUCTION

Beekeepers who operate in short-season areas and/or depend on the rapid build-up of package bees for maximum honey production or to pollinate crops, state that swarming and various types of queen losses usually have an adverse effect on colonies, particularly in terms of honey production.

Farrar and Schaefer (1939) stated that only 49.3 per cent of package queens survive the production season, and only 27.9 per cent survive as good queens. Swarming (Simpson 1959; Free 1967; Farrar 1968) and supersedure (Root and Root 1935; Tontz 1940; Furgala 1962) are factors that adversely affect brood-rearing capacity. During a two-year study of factors affecting honey bee populations at the University of Manitoba, colonies which had queen-related problems, or which swarmed, were compared to normal colonies to ascertain what effects these various problems had on brood rearing and honey production.

MATERIALS AND METHODS

In 1969 and 1970, package bees with queens of a yellow strain were used to initiate colonies of various weights (0.45, 0.9, 1.4 and 1.8 kg) in hives of the Langstroth type (Dadant 1975). Groups of 8-10 colonies of each size were established and managed for honey production. At 19 days after hiving and at 12-day intervals thereafter, the area of sealed brood in each colony was recorded by the methods of Smirl (1970) and Nelson (1971). The period of nectar flow was determined by placing at least one colony of each of the four package weights on a platform scale and recording the daily losses or gains. The average gain for a five-day period was calculated and used to represent the flow pattern (Smirl 1970). The net gain in weight of the honey supers, over the season, was taken as the amount of honey produced. Colonies of each group showing no queen or swarming problems were considered "normal" colonies. "Problem" colonies were categorized as follows:

1. Queen loss – the sudden loss or disappearance of a queen through nonacceptance or through accidents relating directly or indirectly to colony manipulations. This

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Table 1. The amount of sealed brood and honey produced by normal colonies of honey bees and by colonies with queen-related problems or those from which bees swarmed.

Colony identity and number	Year	Package weight (kg)[1]	Occurrence of problem (days from hiving)	Brood production		Honey production		Remarks: problem colonies
				Area (cm^2)[2]	Percent of normal	Weight (kg)	Percent of normal	
				QUEEN LOSS				
1 a[3](1)[4]	1970	0.45	26	15,500	82.9	53.1	52.3	Replaced queen within 6 days.
b(7)				18,692		101.6 ± 7.5[5]		
2 a(1)	1970	0.9	63	15,273	76.4	90.3	69.6	Reared own queen.
b(4)				19,995		129.7 ± 15.2		
3 a(1)	1969	0.9	90	19,931	89.4	26.3	62.3	No queen reared.
b(7)				22,291		42.2 ± 5.0		
4 a(1)	1969	1.4	54	11,610	51.6	12.7	29.5	Queen cells introduced; no queen reared.
b(6)				22,510		43.1 ± 4.6		
5 a(1)	1969	0.9	19	22,368	89.9	65.3	100.6	Replaced queen within 6 days.
b(5)				24,884		64.9 ± 2.1		
6 a(1)	1969	1.4	3;88	20,801	85.2	28.1	69.0	Replaced queen 1 within 6 days; queen 2 not replaced.
b(4)				24,400		42.2 ± 8.9		
7 a(1)	1969	0.9	56;76	16,622	66.8	26.3	40.5	Replaced queen within 6 days.
b(5)				24,884		64.9 ± 2.1		
				QUEEN FAILURE				
8 a(1)	1970	0.9	38	12,985	64.9	83.5	64.4	Same queen all season.
b(4)				19,995		129.7 ± 15.2		
9 a(1)	1970	1.8	75	19,066	88.5	55.3	46.0	Same queen all season.
b(3)				21,536		120.3 ± 11.8		
10 a(1)	1970	1.4	13	18,275	82.5	76.2	58.1	Superseded.
b(5)				22,149		131.1 ± 8.7		
11 a(1)	1970	0.45	68	15,692	83.8	94.4	92.4	Superseded.
b(7)				18,725		102.1 ± 7.5		
12 a(1)	1969	1.4	54	19,602	87.1	29.9	69.4	Same queen all season.
b(6)				22,510		43.1 ± 4.6		
				SWARMING				
13 a(1)	1969	1.4	80	26,303	107.5	19.1	42.9	Swarm caught and returned.
b(6)				24,459		44.5 ± 8.2		
14 a(1)	1969	1.4	92	26,858	106.9	36.3	67.2	Swarm caught and returned.
b(6)				25,123		54.0 ± 5.4		
15 a(1)	1970	1.4	75	19,780	88.6	61.7	47.1	Reared own queen.
b(5)				22,317		131.1 ± 8.7		
16 a(1)	1969	0.9	80	22,652	100.6	15.4	36.5	No queen reared.
b(7)				22,504		42.2 ± 2.4		

[1] 0.45 kg package contains about 3,500 bees.
[2] approximately four cells/cm^2; 12 day measurements upon request.
[3] a = problem colony; b= normal colony.
[4] number of colonies.
[5] mean and standard error.

Table 2. Average amount of sealed brood and honey produced by normal colonies and colonies with queen-related problems or those from which bees swarmed (summary of Table 1).

	Brood production		Honey production	
Type of Colony	Area (cm^2)	Percent of normal	Weight (kg)	Percent of normal
Queen loss	17,444	77.4	43.2	61.9
Normal	22,522		69.8	
Queen failure	17,124	81.6	67.9	64.5
Normal	20,983		105.2	
Swarming	23,898	101.3	31.1	45.8
Normal	23,600		67.9	

loss was usually detected and a replacement queen introduced within six days; after July 1, a colony with a queen loss was usually left to rear its own queen.

2. Queen failure and/or supersedure – the inability of a queen to maintain an egg-laying rate comparable to that in normal colonies; this was usually evidenced by a gradual decline in brood production. In some cases, queen failure resulted in a colony superseding its queen, i.e. the natural replacement of a queen by the colony.
3. Swarming – the natural method of colony reproduction in which the queen and a large number of the workers depart suddenly from the hive and seek a new home. Swarms were detected in or near the apiary site and subsequently by the presence of newly-emerged queens, empty queen cells, or an abrupt decrease in population of the colony.

RESULTS AND DISCUSSION

In colonies with queen loss, which occurred from 3 to 90 days after hiving, the amount of sealed brood produced was less that that of normal colonies (Tables 1 and 2). Brood rearing was least affected where queens were replaced early in the season. Colony 5a shows that when a queen loss occurred early in the season and the queen was quickly replaced, the colony produced a normal crop of honey. Some queenless colonies (3a and 4a) did not rear new queens (Table 1), but this may have been due to disturbances during the collection of data. However, on average, for colonies that lost their queens, brood production was only 77.4% of normal and honey production 61.9 of normal (Table 2).

In colonies with failing queens and/or supersedures, which occurred 13 to 68 days after hiving, the amount of sealed brood and honey produced was less than that produced by normal colonies and similar to that of colonies with queen loss. Brood and honey production in colonies with failing queens were 81.6 and 64.5% of normal, respectively (Table 2).

Colonies that swarmed had more sealed brood prior to swarming than did normal colonies, but in most cases brood production declined below the normal colony level after swarming even if the swarm was returned and the colony remained queenright. Over the season, swarming colonies produced 101.3% of normal (Table 2). However, up to day 75 (the day the first colony swarmed) the sealed brood area of the colonies which swarmed averaged 16% more than did the normal colonies. This is consistent with the findings of Holzberlein (1952), i.e. that colony strength attained early in the season was likely to induce swarming. Honey production, however, was only 45.8% of normal. A 50% reduction in honey yield as a result of swarming has been reported previously (Wulfrath 1957; Babich 1962; Tsibul'skii 1966).

In other studies with package bees, queen-related and swarming problems have been noted in 20 to 45% of colonies (Nelson unpublished). In general, queen-related problems were more prevalent than was swarming.

The nectar flows varied considerably in 1969 and 1970 as indicated by scale colony gains. In 1969, the average gain was 56 kg, with the period of nectar flow occurring between 8 July and 16 September, while in 1970 the average gain was 132 kg, with the period of nectar flow occurring between 3 July and 22 August. In general, there was a greater loss of honey crop in 1970 than in 1969 because of queen-related and swarming problems. This may indicate that a greater percentage loss in honey production occurs during years of high nectar yields, as in 1970, than in years of low nectar yields as in 1969.

The above data show the importance of maintaining colonies with good queens and the need to prevent swarming. If the potential loss of production from the three types of problems discussed in considered, it is evident that the economic losses for a beekeeper could be great. The loss of production from seven colonies that lost their queen was 187 kg; from 5 colonies with queen failure and/or supersedure 187 kg; and from 4 colonies that swarmed 138 kg; a total of 513 kg from 16 colonies. The suggestion of Farrar (1968) that replacement queens be maintained in nuclei (miniature hives with a small population of worker bees used for holding or rearing queens) for rapid replacement of lost or failing queens, seems valid under Manitoba conditions. In years when many queen problems occur, the additional honey produced would more than justify the expense of maintaining nuclei and queens, while in years with few queen problems, the brood and bees from nuclei could be used to strengthen colonies.

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INSECT PARASITES REARED FROM BERTHA ARMYWORM, *MAMESTRA CONFIGURATA* WALKER, COLLECTED FROM ARTIFICIAL FIELD POPULATIONS NEAR WINNIPEG, MANITOBA[1]

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ABSTRACT: Five species of parasitic Hymenoptera, four species of parasitic Diptera and two phorid species that may be parasitic emerged from larvae and pupae of bertha armyworm, *Mamestra configurata* Walk., in artificial infestations on rapeseed near Winnipeg, Manitoba, in 1974-77. Nine of the eleven species are not known to parasitize bertha armyworm in natural infestations; evidence is provided that the present records reflect artificialities in the host populations. Details of the life history and biology of several of the parasitic species are presented.

INTRODUCTION

Larvae of the bertha armyworm, *Mamestra configurata* Walk. have been collected throughout the main rape-growing areas of Manitoba west and north of Winnipeg (Wylie and Bucher 1977). None have been found south of Winnipeg, though small numbers of adults have been trapped there for the past three years (G. L. Ayre, Research Station, Winnipeg, pers. comm.). During 1974-77, larvae and pupae of this species were released at various times in field plots of rape at Glenlea, about 20 km south of Winnipeg, to study dispersal, diapause and damage assessment. As a result of these releases, larvae and pupae were unusually abundant in this locality and, in 1976, were present unusually late in the summer. It seemed possible that the larvae and pupae in these artificial populations would be parasitized not only by species that usually parasitize bertha armyworm (Wylie and Bucher 1977) but by other species as well. Therefore, samples of the larvae and pupae were collected and reared in the laboratory. The rearing results and the results of additional laboratory experiments to obtain details of the biology and life history of some of the parasites follow.

MATERIALS AND METHODS

Seven tests involving releases and collections of bertha armyworm were made (Table 1). In Test 1, non-diapausing pupae were released on a layer of vermiculite moistened with distilled water in wooden boxes which were partly buried in the soil in the rape plots. A wire screen over each box excluded mammals but not insects, and tree branches were placed on the screen to reduce the intensity of sunlight inside the box. Pupae from which moths did not emerge were subsequently removed from the boxes and were reared to determine if they had been parasitized. In Tests 2, 3 and 4, larvae of *M. configurata* were released. The release site in Test 2 was a rape plot surrounded by an electric barrier which limited dispersal of the larvae, and those in Tests 3 and 4 had no barriers. Some of the larvae in each of these three tests were collected later and reared. In Tests 5, 6 and 7, the larvae were released into cages erected over rape plants; the cages were approximately 122 cm high by 91.44 cm square and were of 32-mesh (per in) saran screen which limited dispersal of the larvae. Pupae of *M. configurata* were later removed from the soil inside the cages and reared. All collected hosts were reared at 20°C with a 16 h : 8 h L:D cycle; subsequent laboratory tests were at this temperature and light regime unless indicated otherwise.

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Table 1. Releases and collections of larvae (L) and pupae (P) of *Mamestra configurata* Walker at Glenlea, Manitoba, 1974-77.

Test	Release site	Stage released	Number released	Date of release	Stage collected	Date of collection
1	Boxes in rape plot	P	10,000	early July 1974	P	10 July 1974
2	Rape plot with barrier	L_1	45,000	5 July 1976	$L_5 + L_6$	28 July 1976
3	Rape plot without barrier	$L_1 + L_2$	5,500	23 July 1976	L_6	12 August 1976
4	Rape plot without barrier	$L_1 + L_2$	3,000	4 August 1976	L_6	23 August 1976
5	Screen cage	L_2	400	5 July 1976	P	16 August 1976
6	Screen cage	L_2	400	12 July 1976	P	17 August 1976
7	Screen cage	L_1	500	17 June 1977	P	8 August 1976

RESULTS AND DISCUSSION

Eleven species of parasites emerged from larvae and pupae of *M. configurata* (Table 2). Details for each species follow.

Banchus flavescens Cress. Two parasite larvae emerged in the laboratory on 4 August 1976 and 11 August 1976 from bertha armyworm larvae collected on 28 July 1976. Another parasitized larva of *M. configurata* was collected on 9 August 1974 in an open

Table 2. Insect parasites reared from larvae (L) and pupae (P) of *Mamestra configurata* Walker collected at Glenlea, Manitoba, 1974-77.

Species	Test number	Reared from	No. of parasitized hosts
Hymenoptera			
Ichneumonidae			
Banchus flavescens Cress.	1, 2	L	2
Ichneumon canadensis Cress.	5	P	4
Melanichneumon (Vulgichneumon) brevicinctor (Say)	5, 6	P	3
Netelia sp.	2, 3, 4	L	3
Pteromalidae			
Eupteromalus americanus Gahan	5	P	8
Diptera			
Phoridae			
Diplonevra funebris Mg.	1, 5	P	2
Megaselia sp.	7	P	1
Tachinidae			
Athrycia cinerea (Coq.)	2, 3, 4	L	29
Blondelia sp.	3	L	1
Lespesia archippivora (Riley)	2, 4	L	8
Winthemia rufopicta (Bigot)	4	L	1

rape plot at Glenlea; the parasite larva emerged in the laboratory on 17 August. As no bertha armyworm larvae were released in 1974 the host could have been from an endemic population or from the progeny of a moth produced by pupae released in Test 1. *B. flavescens* parasitizes first, second and third instars of *M. configurata,* matures in the sixth instar and forms a cocoon in the soil (Wylie 1977a).

Ichneumon canadensis Cress. Two males and two females of *I. canadensis* emerged during 20 August to 8 September 1976 from pupae collected on 16 August in the soil beneath a field cage. Wylie and Bracken (1977) reared this pupal parasite from bertha armyworm collected in a field cage at Grosse Isle, Manitoba, described details of its biology and life history and concluded that it occasionally parasitizes *M. configurata* in nature.

Melanichneumon (Vulgichneumon) brevicinctor (Say). One male and two females of this ichneumonid emerged during 31 August to 5 September 1976 from bertha armyworm pupae collected on 16 and 17 August in the soil beneath a field cage. As this was the first time that *M. (V.) brevicinctor* had been reared from *M. configurata* and as little was known about it, laboratory studies were made of its biology and life history. Females of *M. (V.) brevicinctor* mated within 24 h of emerging and parasitized bertha armyworm when less than 24 h old. They attacked both male and female pupae, and diapausing and non-diapausing pupae, and showed no preference for 1-day old, 7-day old or 14-day old pupae. They did not parasitize sixth instars of *M. configurata.* The parasite females drilled and oviposited into both parasitized and unparasitized hosts. After withdrawing the ovipositor from the pupae, females of *M. (V.) brevicinctor* usually fed on the host's fluids that accumulated on the surface at the drilling site. Up to six parasite larvae per host were dissected from bertha armyworm pupae parasitized in the laboratory. Combat among the first instars eliminated all but one parasite in each superparasitized host. Development time from oviposition to adult emergence was about 16 days for males and 17 days for females at 25°C, 22 and 23 days at 20°C, and 44 and 46 days at 15°C. Thirty-nine of 86 parasite progeny propagated in the laboratory were females. Although some females of *M. (V.) brevicinctor* lived nearly 6 months at 15°C, they probably do not hibernate, for females could not fly and did not attempt to drill at 25°C if previously chilled at -5°C for one week. The overwintering stage is unknown. *M. (V.) brevicinctor* probably has two or more generations each year in Manitoba, for I was able to propagate it without diapause on *M. configurata.* I also propagated the parasite on pupae of variegated cutworm, *Peridroma saucia* (Hubn.) (Lepidoptera: Noctuidae); and Heinrich (1961) lists other noctuids as well as arctiids, geometrids and pyralids as hosts.

Netelia sp. One sixth instar of bertha armyworm with an ectoparasitic larva was collected on 12 August 1976. The parasite spun a cocoon and an adult subsequently identified as *Netelia* sp. emerged 1 September. Two other sixth instars of *M. configurata* collected in 1976 had been parasitized, probably by *Netelia* sp. One, collected 28 July, had an ectoparasitic larva similar to *Netelia* sp. as well as two macrotype tachinid eggs. The host died before the parasites matured. The second bertha armyworm had a black hymenopterous egg on the first abdominal segment when collected on 23 August. The larva that hatched from the egg was similar to those on the other two hosts; it died before maturing, probably from premature death of the host which produced seven puparia of *Lespesia archippivora* (Riley). The parasite egg and larva closely resembled those of *Paniscus* sp. described by Strickland (1923). *Paniscus* is no longer a valid genus and most of its species have been reassigned to *Netelia.* On this basis, I believe that both of the unidentified parasite larvae pertain to the genus *Netelia.* Species of the genus *Netelia* parasitize various Lepidoptera including noctuids (Guppy 1961; Schaaf 1972) but none has been reared previously from *M. configurata.*

Eupteromalus americanus Gahan. Up to 100 adults of this pteromalid emerged during 30 August to 13 September 1976 from each of eight bertha armyworm pupae collected on 16 August in the soil beneath a field cage. The adults mated within 48 h of emergence. Twenty parasite females drilled into and fed on 56 non-diapausing pupae of *M. configurata* that were exposed to them in the laboratory; but no parasite eggs were found in six of the hosts that were dissected, and no parasite progeny emerged from the others. No dead

immature parasites were found when host pupae that died in this test were dissected. As no other pupae of bertha armyworm were available for study, the reason why *M. configurata* was not parasitized in the laboratory test is unknown. Females of *E. americanus* attempted to drill into cocoons of *B. flavescens* but failed to pierce the cocoons. Bertha armyworm is the first lepidopterous host recorded for *E. americanus.* I have also reared *E. americanus* from a puparium, probably of a sarcophagid, collected 28 September 1976 in leaf litter in a stand of mixed hardwood trees at Glenlea. Peck (1963) listed puparia of the Hessian Fly, *Mayetiola destructor* (Say) (=*Phytophaga destructor* (Say)) (Diptera: Cecidomyiidae) as a host of *E. americanus.*

Diplonevra funebris Mg. Ten adults of this phorid emerged 16 August 1974 from a bertha armyworm pupa that had been placed in the field in early July and returned to the laboratory on 10 July. In addition, one adult of *D. funebris* emerged in the laboratory on 2 September 1976 from a bertha armyworm pupa collected in the soil beneath a field cage on 16 August. It is not clear whether the larvae of *D. funebris* are parasitic or saprophagous. R. V. Peterson (Biosystematics Research Institute, Ottawa; pers. comm.) indicates that little is known of the species or genus.

Megaselia sp. Adults of *Megaselia* sp. emerged in August 1977 from a pupa of *M. configurata* collected on 8 August in the soil beneath a field cage. This phorid may be parasitic (R. V. Peterson, Biosystematics Research Institute, Ottawa; pers. comm.). The various subgenera of the genus *Megaselia* include species with a wide range of feeding habits (Cole 1969).

Athrycia cinerea (Coq.) Twenty-nine of the larvae of *M. configurata* collected during late July to 23 August 1976 produced 47 puparia of *A. cinerea* (range 1-4 per larva). Thirty-eight of the parasite pupae went into diapause, seven developed without diapause and two could not be classified because they died soon after the puparia formed. Five of the hosts had been multiparasitized: three by *L. archippivora* (two, one and one adults of this tachinid emerged from each host); one by *Blondelia* sp. (one adult); and one by *Winthemia rufopicta* (Bigot) (one adult). *A. cinerea* is a widely distributed, common larval parasite of *M. configurata* in the prairie provinces (Wylie and Bucher 1977). Details of its biology, life history and diapause were published by Wylie (1977a, b).

Blondelia sp. One adult of this tachinid emerged on 30 August 1976 from a larva of *M. configurata* collected on 12 August. The host also produced a puparium of *A. cinerea.*

Lespesia archippivora (Riley). One bertha armyworm larva collected on 28 July 1976 and seven others collected on 23 August 1976 were parasitized by this tachinid. Parasite larvae from one host formed puparia in early August and from the others during 28 August to 13 September, and parasite adults emerged on 15 August and during 13 September to 30 September. The eight hosts contained a total of 16 puparia of *L. archippivora* (range 1-7). Three of the hosts collected on 23 August also produced puparia of *A. cinerea,* and one had a hymenopterous ectoparasite which died before maturing and was tentatively identified as *Netelia* sp. This is the first published report of this tachinid parasitizing *M. configurata. L. archippivora* is widely distributed in Canada and the United States and parasitizes larvae of noctuids (Breeland 1958; Harding 1976) and nymphalids (Cole 1969). Adults were collected on numerous occasions during 5 July to 13 August 1974 in light traps at Swan River, Manitoba, but none was reared from larvae of *M. configurata* collected at that time in adjacent fields.

Winthemia rufopicta (Bigot). One bertha armyworm larva collected on 23 August 1976 produced one puparium of this tachinid from which an adult emerged on 15 September. The host also produced four puparia of *A. cinerea.* There are no published reports of this species parasitizing *M. configurata.* I have also reared *W. rufopicta* from an arctiid larva, tentatively identified as *Isia isabella* (J. E. Smith) collected at Roblin, Manitoba on 4 August 1976; and from a larva of the armyworm, *Pseudaletia unipuncta* (Haw.) collected 12 July 1977 at Elma, Manitoba. Danks (1975) reared *W. rufopicta* from larvae of *Heliothis zea* (Boddie) and *H. virescens* (Fab.) (Lepidoptera: Noctuidae) in North Carolina.

CONCLUSIONS

Only two of the eleven species, *B. flavescens* and *A. cinerea,* are know to parasitize *M. configurata* in natural infestations (Wylie and Bucher 1977). Probably some of the other nine species were recorded in the present study because of artificialities in the host populations. For example, high host larval densities could have been responsible in some of the tests, especially Test 2 where dispersal of the larvae was limited. Wylie and Bucher (1977) reared small numbers of several parasite species (other than the 11 species reported in this study) from *M. configurata* in natural infestations in 1972 and 1973 when host densities were high, but none in subsequent years when host numbers were lower. Second, late occurrence of the host larvae could have been responsible in Test 4 (Table I): first and second instars were released on 4 August when third instars or larger larvae are present in natural populations; and mature larvae were collected on 23 August when most larvae in natural populations have already pupated. Because they were present later than usual, the hosts might have been exposed to parasite species with which they are not usually synchronized temporally. Third, the cages in Tests 5, 6 and 7 could have increased the probability of parasitism by the larger pupal parasites (*I. canadensis* and *M. (V.) brevicinctor*) because the fine mesh screen would have prevented escape of any adults of these species that were trapped when the cages were erected or that emerged later from the soil inside the cages.

Although these characteristics of our artificial populations could have increased the probability of parasitism, I suspect that most of the species occasionally parasitize bertha armyworm in natural infestations. For example, *Netelia* sp., *L. archippivora* and *W. rufopicta* parasitize various other noctuids; and species of the genus *Blondelia* have been reared from Lepidoptera in several families (Cole 1969). The pupal parasites would have been missed in the earlier study (Wylie and Bucher 1977) in which the hosts were collected after 11 September, i.e. later than the pupal parasites emerged in the present study. Most of the nine incidental parasites are known to be non-host specific and probably have little impact on bertha armyworm abundance.

Because natural populations of larvae of *M. configurata* are unknown at Glenlea, I suspect that the two main bertha armyworm parasites, *B. flavescens* and *A. cinerea* develop on other host species in this locality. Additional information on other hosts for these two parasites elsewhere in Manitoba will be published separately.

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PHOTOPERIODIC AND TEMPERATURE REGULATION OF DIAPAUSE INDUCTION IN THE BERTHA ARMYWORM, *MAMESTRA CONFIGURATA* WALKER[1]

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ABSTRACT: The bertha armyworm, *Mamestra configurata* Walker, has a facultative pupal diapause induced by photoperiods of 5 to 13 h at 20°C. The critical photoperiod at this temperature was 14.75 h. At 23°C the critical photoperiod was decreased by about 1 h. At 17°C diapause was induced even under non-diapause photoperiodic conditions whereas at 27°C diapause was completely inhibited under the diapause-inducing photoperiods studied. Larvae from the 2nd to the early 6th instar were sensitive to factors that induced or averted diapause. The reciprocal transfer of larvae from short or long photoperiods or high or low temperatures, at different stages of development, indicated that the critical number of short-day or high-temperature cycles needed was about 13 to 15 and the critical number of short-day or low-temperature cycles needed was about 8 to 9.

INTRODUCTION

The bertha armyworm, *Mamestra configurata* Walker, is native to the Canadian prairies and parkbelt region (Rempel 1951) and is an important agricultural pest of cruciferous crops. According to King (1928, 1929) a widespread outbreak of this insect in 1927 and 1928 covered Manitoba, Saskatchewan and Alberta and even extended into British Columbia. An outbreak in 1971 and 1972 caused extensive damage to rape, *Brassica compestris* L. and *B. napus* L. Putnam (1972) gives a brief life history of this insect. The larvae mature in late August and early September and enter the soil to pupate. The moths emerge in mid June and early July of the following year. In the Canadian prairies the bertha armyworm apparently has one generation a year, but fall flights of moths have been observed (King 1928) indicating that diapause induction in such instances was incomplete. This study was undertaken to provide more precise knowledge on the photoperiodic and temperature requirements for diapause induction and the sensitivity of developmental stages to diapause-inducing conditions.

METHODS AND RESULTS

The bertha armyworms used in this study were taken from a stock culture maintained at the Winnipeg Research Station. They were reared on a semi-defined diet as described by Bucher and Bracken (1976) except for special photoperiod and temperature conditions described in the text.

Immediately after pupation the pupae were transferred to a rearing cabinet at 25°C. Diapause and non-diapause pupae were distinguished on the basis of one or more of the following criteria: the presence or absence of eye pigmentation in 10-12 day old pupae and the duration of pharate adult development. In non-diapause pupae, visible black pigmentation appeared in the compound eye of developing pharate adults within 10-12 days after pupation and the pharate development was completed in about 25 days.

Photoperiod and Diapause Incidence

Bertha armyworm larvae were reared after hatching at 20 ± 0.5°C at daily photoperiods of 0, 1, 5, 9, 10, 11, 12, 13, 14, 15, 16, 18 and 24 h. Each experiment was

[1] Contribution No. 760, Agriculture Canada, Research Station, Winnipeg, Manitoba.

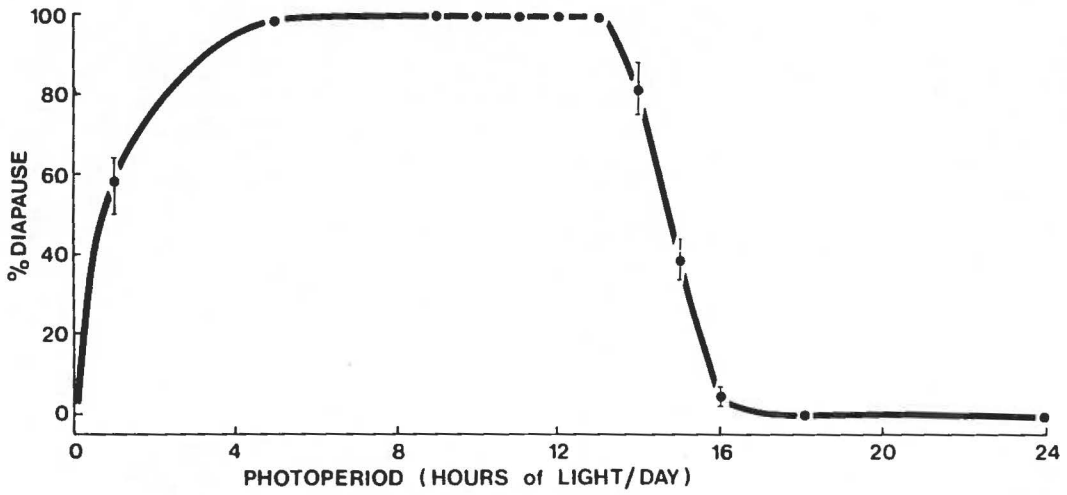

Figure 1. Effect of photoperiod at 20°C on the incidence of diapause in *M. configurata.* Mean (± S.E.) of 3 determinations.

replicated three times (N = 50-60 insects/replicate). Photoperiods ranging from 5 to 13 h induced 100% diapause (Figure 1). From the graph it may be seen that the critical photoperiod (the photoperiod required to induce diapause in 50% of the test population) in the long-day regimen was around 14.75 h. A low incidence of diapause occurred at the 0 h and 16 h photoperiods. Photoperiods above 16 h did not induce diapause. In the short-day regimen, 1 h of light per day induced diapause in about 50% of the individuals.

Temperature and Diapause Incidence

Insects were reared at daily photoperiods ranging from 12 to 18 h at temperatures of 17, 20, 23 and 25°C. Each experiment was replicated three times (N = 50-60 insects/replicate). The mortality was low (< 9%) and the results have been expressed as per cent of total surviving insects. At 17°C, diapause occurred in all the insects regardless of photoperiod (Figure 2). At 25°C no diapause occurred in photoperiods of 14 to 18 h/day and less than 10% diapause occurred at photoperiods of 12 and 13 h/day. An increase of 3°C in rearing temperature (from 20 to 23°C) decreased the critical photoperiod for diapause induction by about 1h (Figure 2).

Sensitivity of Developmental Stages

Four different photoperiod-temperature dependent regimes were selected, two of which induced diapause and two of which averted diapause when larvae were reared from first instar to pupation. The diapause-inducing regimes were LD 12:12 (light-dark cycle/24 h period) at 20°C (photoperiod induced diapause) and LD 18:6 at 17°C (temperature induced diapause). The diapause-averting regimes were LD 18:6 at 20°C (photoperiod induced non-diapause) and LD 12:12 at 27°C (temperature induced non-diapause). For determining the sensitive period for diapause induction and the effect of subsequent exposure to non-diapause conditions, the larvae were transferred from the diapause regime of LD 12:12 at 20°C to the two non-diapause regimes of LD 18:6 at 20°C and LD 12:12 at 27°C. The larvae were transferred at an interval of 5 days starting soon after hatching. Transfers from non-diapause regime of LD 18:6 at 20°C to diapause-inducing conditions of LD 12:12 at 20°C and LD 18:6 at 17°C were also made.

The duration of each instar at 20°C is indicated in Figure 3a (L_1 to L_6 indicating the 6 instars, L_6 taken to include prepupal stage). Transfer of larvae during the first instar from diapause condition to non-diapause condition prevented diapause (Figure 3c). Transfer of later instars in this fashion resulted in progressively higher diapause incidence. A 50% diapause incidence occurred when larvae were transferred about the middle of the fourth instar (16 days). This diapause-inducing effect of short-day photoperiod could be reversed

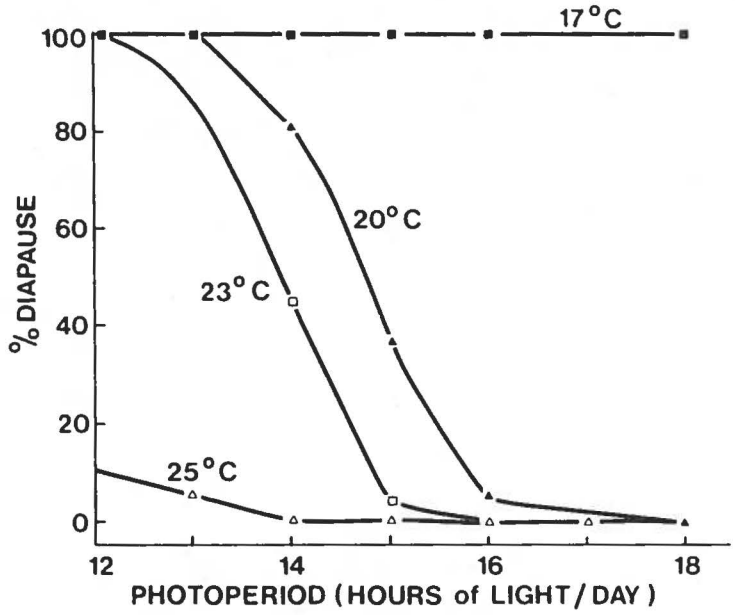

Figure 2. Effect of temperature on photoperiodic induction of diapause in *M. configurata*. Each point on the graph represents an average of 3 determinations.

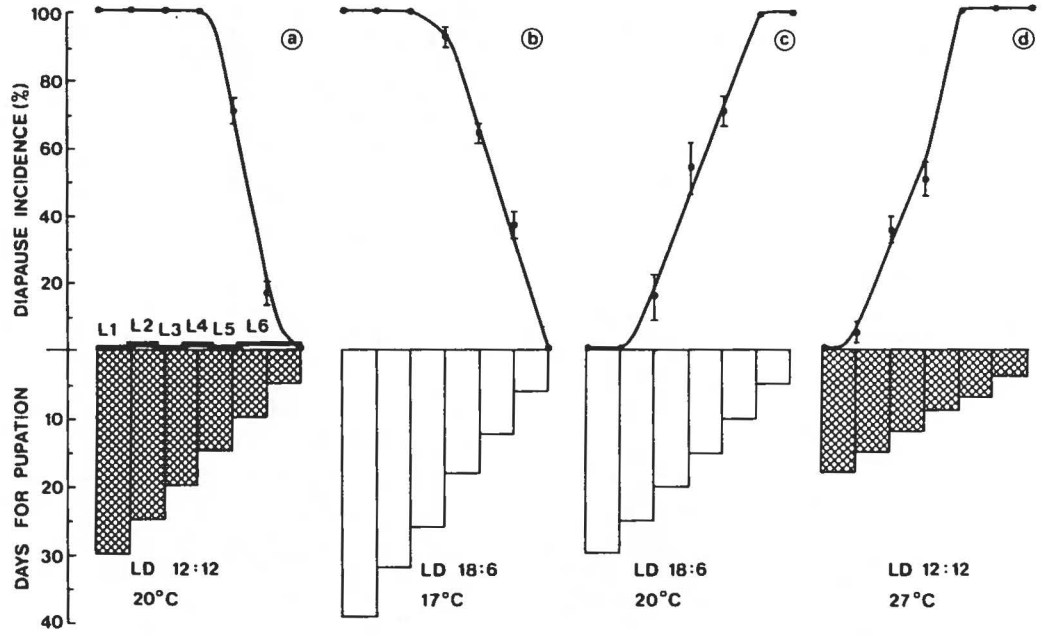

Figure 3. Effects on *M. configurata* of conditions that induce or avert diapause. Duration of larval development (instars L_1 to L_6 and prepupa) shown on abscissa. Larvae transferred at 5-day intervals, from hatching, from non-diapause (LD 18:6, 20°C) to diapause regimes (a,b) and from diapause (LD 12:12, 20°C) to non-diapause regimes (c,d). Mean (± S.E.) of 3 to 5 determinations.

in at least 1/3 of the population as late as the mid fifth instar. It was only sometime during the last instar that it was irreversibly fixed. The photoperiod-induced diapause, however, was more resistant to reversal by temperature (Figure 3d). It seems to be irreversibly fixed by the middle of the fifth instar in all individuals. The 50% level of diapause sensitivity remained at mid fourth instar as in the previous case.

When larvae were transferred from photoperiod-induced non-diapause conditions to photoperiod-induced diapause conditions (Figure 3a) the larvae were sensitive until about the mid fourth instar after which the sensitivity for diapause induction decreased rapidly. A 50% diapause incidence may still be obtained with early sixth instar larvae. With temperature as the factor inducing diapause only the first three instars were sensitive (Figure 3b). The fall in sensitivity was gradual for the rest of the instars. The 50% level, however, did not seem to change from the previous case.

DISCUSSION

It is evident from these studies that the photoperiodic response curve in the bertha armyworm is of the long-day type with a facultative diapause. At 20°C, diapause (100%) was induced when larvae were reared at photoperiods of 5 to 13 h. Longer photoperiods at this temperature averted diapause. Temperature influenced the range of inductive photoperiods. Low temperatures (17°C) induced diapause irrespective to photoperiodic regimes and high temperature (27°C) averted diapause. Thus, the diapause in the bertha armyworm resembles that of many other insects which have evolved in temperate regions where they are subjected to marked seasonal changes in photoperiod and temperature (Danilevskii 1965).

The most important feature of the photoperiodic response is the critical day length which separates the long photoperiods resulting in non-diapause development from the short photoperiods which ultimately lead to the dormant state. Danilevskii (1965) has pointed out that a photoperiodic response curve includes responses at both natural and unnatural photoperiods. Those towards the right hand side of the curve (Figure 1) represent those day lengths which occur normally during the part of the year when temperature and other climatic factors are suitable for insect development. The proportion of the pupae entering diapause falls off in ultrashort ($<$ 1 h) day lengths. This type of photoperiodic response is similar to that observed in *Pectinophora gossypiella* (Saunders) (Pittendrigh and Minis 1971), *Ostrinia nubilalis* (Hbn) (Beck and Hanec 1960) and *Pieris brassicae* (L.) (Danilevskii 1965).

Constant temperatures are also known to affect the critical day lengths in a number of long-day species. The species may show a temperature compensated response as in *P. brassicae* (Danilevskii 1965) and *O. nubilalis* (Beck and Hanec 1960) or the critical day length may decrease steadily as the temperature rises as in *Acronycta rumicis* (L.) (Danilevskii 1965). The bertha armyworm differs slightly from the latter group. A temperature change of 3°C, i.e. from 20°C to 23°C, reduced the critical photoperiod by almost 1 h, but at 27°C (LD 12:12) no diapause was induced. Similarly, at 17°C (LD 18:6) diapause induction was total.

The modifying effect of temperature on the photoperiodic response is known in many long-day species. In *P. brassicae,* for example, a short-day (LD 12:12) is fully inductive up to about 25°C. At higher temperatures the proportion of larvae entering diapause drops sharply so that at about 30°C no diapause is induced (Danilevskii 1965). Pupal diapause in the tomato moth, *Diataraxia oleracea* L. is induced in short-days at temperatures below 30°C; the incidence drops above this temperature (Way and Hopkins 1950). In a natural environment the low temperature limit of photoperiodic reaction may be of some ecological importance for *M. configurata* and doubtless is an adaptation to the cold autumn conditions of the northern regions. In the prairie provinces most larvae pupate between mid August and mid September in an average year.

The reciprocal transfer of insects from long- to short-day cycles during development has been used to map out the sensitive period and to determine the inductive cycles required for diapause. In Lepidoptera, the greatest sensitivity to photoperiod conditions

is shown by the larvae. Tanaka (1950), working with the pupal diapause in *Antheraea pernyi* Guerin, transferred the larvae from long-days (LD 18:6) to short-days (LD 9:15) and vice versa at different stages of development and found that the sensitive stage extended at least back to the second instar. In *M. brassicae* (L.), sensitivity remained uniform throughout larval development and the percentage of diapausing larvae increased in proportion to the duration of action of the short-day (Masaki 1957). In *M. configurata* diapause was induced either by varying the temperature or photoperiod. At 20°C, long days averted diapause and short days induced diapause. However, diapause was averted independent of photoperiod at 27 and 17°C respectively. The sensitivity of the reaction to factors that induce or avert diapause was shown by second to early last-instar larvae. The transfer of larvae from short to long photoperiods or from low to high temperatures at different stages of development indicated that the critical number of short-day or high-temperature cycles (i.e. the number of daily cycles needed to produce 50% diapause) was about 13 to 15 (Figure 3c, d). The early to mid fourth instar is considered to be a critical stage. When the transfer was from long to short photoperiod or from high to low temperatures, the critical number of short-day or low-temperature cycles needed was about 8 to 9 (Figure 3a, b) and the early sixth instar may be considered as a critical stage. In nature, the prepupal stage is in the soil about 1 to 2 days before pupation so that the actual number of short-day or low-temperature cycles needed may be less than eight. In *M. configurata,* the diapause promoting effect of short days was "stronger" than diapause inhibiting effects of long days. This kind of response differs from that observed in *A. pernyi* where the inhibition of diapause by long days was stronger (Tanaka 1950).

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MOSQUITO LARVAE: TEST ORGANISMS FOR SCREENING LIQUID EFFLUENTS FOR THEIR POTENTIAL TOXICITY

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ABSTRACT: We suggest that *Aedes aegypti* L. larvae can be used as test organisms to screen liquid effluents for their potential toxicity to aquatic animals. This would reduce the number of concentrations of the effluent which must be included in a standard fish bioassay. Since the apparatus and procedure required to test the toxicity of a liquid effluent to larvae are simpler than those for fish, there would be savings of time and money.

Salts of copper and zinc, and 2, 4-dinitrophenol, are chemicals frequently added to the secondary cooling water of thermal power stations to control corrosion, scaling, and the growth of algae and other organisms. We have determined the LC_{50}'s of $CuSO_4$, $ZnSO_4$ and 2, 4-dinitrophenol for *A. aegypti* larvae and found them comparable to values published in the literature for fish species commonly used in standard bioassay procedures. We also found that the incipient lethal levels of these chemicals for the larvae compared favorably with published values for fish.

INTRODUCTION

Many chemicals enter the aquatic environment via liquid effluents released to rivers and lakes. For example, the chemical composition of heat exchanger cooling water discharged from thermal electricity generating stations is highly variable, and is unique to each station. It depends on such factors as intake water quality, and the additives used for the control of biological growths, corrosion and scaling. Mixtures of chromate, zinc, phosphate, and silicates are frequently used for corrosion control. Chlorine, hypochlorites and non-oxidizing organics, such as dinitrophenols and organometallics, are used to control bacterial growths. Thus, the list of chemicals that might be present in the cooling water discharged from a thermal generating station is a long one.

The generally accepted bioassay methods for liquid effluents, particularly those which use fish as the test organism, are time consuming and usually require sophisticated apparatus. An aquatic organism which is easily maintained and requires simple test apparatus could, therefore, be advantageously used as a test animal for screening liquid effluents for their potential toxicity. Preliminary examination or 'screening' of the effluent for its toxicity would reduce the number of concentrations to be included in a bioassay, and the time required to establish the permissible quantity of chemicals in the effluent that could be released to a river or lake.

Considerable effort has been devoted to finding chemicals which kill mosquito larvae but there are few references in the literature to the use of mosquito larvae for testing the toxicity of liquids. Marcovitch (1928) investigated the relative toxicities of arsenicals and fluorides to *Culex quinquefasciatus* Say larvae. Mosquito larvae and a cladoceran, *Daphnia magna* Straus, were used by Stroganov, *et al.* (1972) in a study of the toxicity of chemical "anti-biodeteriorants" for some aquatic organisms. Guthrie and Wiewel (1977) used *Aedes aegypti* larvae to assess the chronic toxicity of the terphenyl compound used as the primary coolant for the Whiteshell Nuclear Research Establishment's nuclear reactor. They pointed out, however, that the significance of their results was dependent on whether the sensitivity of the larvae to the primary coolant was representative of that of aquatic invertebrates.

This is a report on the toxicity of a rearing solution containing $CuSO_4$, $ZnSO_4$, or 2, 4-dinitrophenol to *Aedes aegypti* larvae. These chemicals (toxicants) are commonly used as additives in the secondary cooling water of nuclear power stations (Becker and Thatcher 1973). We do not suggest that mosquito larvae be used for assessing the sub-lethal effects of a liquid effluent, or for determining its 'safe' concentration for release to the aquatic environment. Our purpose was to determine if *A. aegypti* larvae could serve as a test organism for screening liquid effluents for their potential toxicity, thereby reducing the time and space required for bioassay with fish. To be useful, it follows that the toxicity of a chemical compound for the larvae should be comparable with the toxicity of that compound for the fish species normally used in bioassay, for example, the fathead minnow, *Pimephales promelas* (Rafinesque), and the rainbow trout *Salmo gairdneri* Richardson.

MATERIALS AND METHODS

Test Organism

First instar *Aedes aegypti* larvae were hatched from eggs obtained from the laboratory colony maintained by R. A. Brust, Department of Entomology, University of Manitoba. The hatching and rearing methods were essentially those described by Brust (1968). The larval-rearing media were changed every 48 h, at which time the larvae were fed a measured quantity of liver powder (Nutritional Biochemicals Corp., Cleveland, Ohio, U.S.A.). The rearing media were sampled for toxicant analysis at the beginning and end of each 48 h period.

Treatment Solutions

Stock treatment solutions were prepared by dissolving $CuSO_4$, $ZnSO_4$ or 2, 4-dinitrophenol (DNP) in distilled water. Aliquots of these solutions were diluted with distilled water to provide the various concentrations of toxicant for the rearing media. The nominal and the measured concentrations of the treatment rearing solutions are given in Table 1. The treatment concentration was calculated by taking the mean of the concentrations at the beginning and at the end of each 48 h media change period.

Experimental

Both chamber and bench tests were run. The chamber tests were run at constant temperature and photoperiod. The purpose of the bench test was to determine whether useful estimates of toxicity could be obtained under conditions of fluctuating temperature and photoperiod. This would enhance the utility of mosquito larvae as a screening organism by doing away with the need to rear them in a constant environment.

Groups of 50 newly hatched larvae were randomly assigned to 15 cm diameter glass petri dishes with covers. The dishes contained either distilled water (controls), or a known concentration of toxicant in distilled water (treatments). Each control and treatment was replicated three times and the replicates randomly assigned either to the chamber-test group or to the bench-test group. The chamber tests were run three times, the bench test once. The treatment concentrations (Table 1) were measured by Analytical Science Branch.

The chamber-test larvae were reared in a controlled environment chamber at $25.0 \pm 0.1^{\circ}C$, 80-85% relative humidity, and a 12L:12D photoperiod. The bench-test larval dishes were placed on the laboratory bench under the photoperiod that prevails in southern Manitoba during the months of January to May. The laboratory temperature during this period varied between 17 and $30^{\circ}C$, and averaged $21.8^{\circ}C$. The toxicity of DNP under bench-test conditions was not determined.

The numbers of dead larvae were counted every hour during the first eight hours of the chamber and bench tests. Thereafter, larval mortality, and the numbers of live and dead pupae, were determined every 24 h until pupation was finished. The elapsed times from hatching to start of pupation for the chamber and bench tests averaged 288 h and 336 h, respectively.

Table 1. Nominal and treatment concentrations of toxicants tested.

Toxicant	Nominal concentration (mg/l)	Treatment concentration[1] (mg/l)
$CuSO_4$	Controls	Not detectable ($<<$ 0.01)
	0.02	0.02 ± 0.014
	0.06	0.06 ± 0.001
	0.1	0.09 ± 0.005
	0.2	0.18 ± 0.008
	0.4	0.34 ± 0.021
	1	0.88 ± 0.086
	5	5.15 ± 0.359
$ZnSO_4$	Controls	Not detectable ($<<$ 0.01)
	0.02	$<$0.03
	0.03	0.03 ± 0.005
	0.1	0.09 ± 0.006
	0.2	0.21 ± 0.006
	0.5	0.45 ± 0.007
	1	0.92 ± 0.014
	5	4.76 ± 0.104
	10	9.73 ± 0.118
	50	52.00 ± 0.845
2, 4-dinitrophenol (DNP)	Controls	Not detectable ($<$ 0.02)
	0.05	$<$0.1
	0.1	0.11 ± 0.021
	0.5	0.50 ± 0.038
	1	1.01 ± 0.061
	5	4.97 ± 0.097
	10	9.27 ± 0.546
	20	21.00 ± 0.550

[1] Mean ± 1 S.E. of the toxicant concentrations measured at the beginning and at the end of each 48 h period.

Analysis of Data

The toxicity of the three toxicants, $CuSO_4$, $ZnSO_4$ and DNP, was assessed in terms of the concentration which killed 50% of the larvae during the larval stadia (LC_{50}), and the incipient lethal level (ILL). The ILL is that level of an environmental entity, the concentration of toxicant in this instance, beyond which the organism can no longer survive for an indefinite time (Fry 1947).

The total larval mortality that occurred in each treatment concentration was adjusted for the mortality which occurred in the controls using Abbott's formula (Matsumura 1975). The LC_{50} of each toxicant was then determined by probit analysis (Finney 1971) of the log toxicant concentrations.

ILL's were estimated as follows: The median response-time (ET_{50}) in each concentration of toxicant was determined by probit analysis of the cumulative larval mortality uncorrected for control-deaths. The log ET_{50} for each concentration was then plotted against log concentration of toxicant. The intercept on the log concentration axis of the asymptote parallel to the log ET_{50} axis marks the approximate value of the ILL. Sprague

(1964; 1969) has described the procedure for estimating the ILL. This is a subjective method since it requires visual inspection of a graph, therefore, no statistical confidence limits can be placed on the estimated ILL.

RESULTS

The results of the three chamber-test runs were homogeneous, therefore, the data were pooled (pooled-chamber). The results of the replicates of the bench test also were homogeneous and were pooled (pooled-bench).

LC_{50}

The LC_{50}'s for larvae exposed to $CuSO_4$, $ZnSO_4$ and DNP for the chamber and for the bench tests are given in Table 2 which also includes toxicity values published for chironomid larvae, fathead minnows and rainbow trout. Since pupae are easier to count than larvae, the LC_{50} for each toxicant was also calculated using the difference between the number of newly hatched larvae and the number of larvae that pupated, as the measure of larval mortality. There was no significant ($p < 0.05$, t-test) difference between the LC_{50} of each toxicant calculated from larval mortality (Table 2), and that calculated from pupation results.

Temperature and photoperiod clearly influenced the bench-test LC_{50}'s. The average temperature to which the bench-test larvae were exposed was about 4°C less than that of the chamber-test larvae, and the temperature variation was also much greater. All the bench-LC_{50}'s were consistently higher and their fiducial limits greater than those of the chamber-LC_{50}'s (Table 2). The fiducial limits of the DNP chamber-test and the $ZnSO_4$ bench-test LC_{50}'s could not be calculated because of the large heterogeneity associated with their respective probit analyses.

The toxic action of either $CuSO_4$ or $ZnSO_4$ appeared to affect all larval instars equally. In contrast, the action of DNP appeared to be more of an 'all or none' effect. Compared to $CuSO_4$ and $ZnSO_4$, the majority of the larvae which survived in DNP to the late instar II stage successfully pupated. This observation may demonstrate acclimation to a toxicant, similar to that which has been reported for fish (Sprague 1969).

Incipient Lethal Level (ILL)

As explained under Methods, the ILL was estimated from a graph obtained by plotting the logarithm of ET_{50} (Table 3) against log concentration of toxicant. The estimated ILL's are given in Table 4. The $CuSO_4$ chamber-, and bench-test ILL's were similar, as were those for $ZnSO_4$ (Table 4), and compare favorably with those reported for juvenile salmon by Sprague and Ramsey (1965) who exposed their fish in soft water. Becker and Thatcher (1973) do not report any ILL's for aquatic organisms exposed to DNP.

DISCUSSION

The purpose of this study was to ascertain if mosquito larvae could be used as a test animal to screen liquid effluents for their potential toxicity to aquatic organisms. Our results indicate that a satisfactory estimate of the LC_{50} can be made from the number of larvae which pupate. This is advantageous since pupae are easier to count than larvae. However, it affords a cruder estimate of mortality since no correction is made for 'missing' or 'lost' larvae when only pupae are counted. More importantly, the LC_{50}'s determined for $CuSO_4$, $ZnSO_4$ and DNP are comparable with those obtained using standard bioassay tests (Table 2).

The screening test procedure would be further simplified if it could be run at room temperature. Our results (Table 2) show that the best estimate of LC_{50}, as would be expected, was that obtained under constant temperature and photoperiod. Nevertheless, the bench-test conditions did provide a useful estimate of the LC_{50}.

The ILL is a more sensitive indicator of potential toxicity than is the LC_{50} (Sprague 1969), but it requires cumulative mortality assessment which is a more time-consuming procedure. The chamber-test ILL's for $CuSO_4$ and $ZnSO_4$ are in agreement with those

Table 2. Mortality, expressed as LC_{50}[1], corrected for mortality in controls, of pooled results for *Aedes aegypti* larvae, instars I to IV, reared in various toxicants either in a constant environment (Chamber), or on the laboratory bench (Bench). Also shown are published toxicities of the toxicants for other aquatic organisms.

	Toxicities					
	A. aegypti		Other aquatic organisms[2]			
Toxicant	LC_{50} (mg/1)	95% probability fiducial limits	Test organism	Concentration (mg/1)	Remarks	Reference
$CuSO_4$ (Chamber)	0.12	0.09 - 0.16	Chironomid larvae	10.00	Survived 7 days; soft water	Fisher (1956)
			Pimephales promelas (fathead minnow)	0.05	96 h - TLm[3]; soft water at 25°C	Tarzwell & Henderson (1960)
$CuSO_4$ (Bench)	0.43	0.11 - 1.70	–	–	–	–
$ZnSO_4$ (Chamber)	1.16	0.49 - 2.66	*Daphnia magna*	1.80	Toxicity threshold; 2 days at 23°C	Bringmann & Kühn (1959)
			Pimephales promelas (fathead minnow)	0.78-0.96	96 h - TLm; soft water at 25°C	Pickering & Henderson (1964)
$ZnSO_4$ (Bench)	$>2;<4$	–	–	–	–	–
DNP (Chamber)	$>5;<10$	–	"sewage organism"	100.00	50% inhibition of BOD	Hermann (1959)
			Salmo gairdneri (rainbow trout)	10.00	Dead, 2.5-6.5 h; 10°C	MacPhee & Ruelle (1969)

[1] LC_{50} - concentration which killed 50% of the larvae.

[2] Published values.

[3] TLm - median tolerance limit.

Table 3. Mortality, expressed as ET_{50}[1], uncorrected for mortality in controls, of pooled results for *Aedes aegypti* larvae, instars I to IV, reared in various concentrations (Table 1) of $CuSO_4$, $ZnSO_4$, or 2,4-dinitrophenol (DNP) either in a constant environment (Chamber) or on the laboratory bench (Bench).

Nominal toxicant concentration (mg/l)	Chamber		Bench	
	ET_{50} (hours)	95% probability fiducial limits	ET_{50} (hours)	95% probability fiducial limits
$CuSO_4$				
≤0.02	A[2]	–	A	–,
0.06	452	390 - 587	982	747 - 1502
0.1	266	243 - 301	–	–
0.2	106	68 - 183	264	241 - 322
0.4	105	65 - 130	150	110 - 195
1	45	39 - 51	75	60 - 105
5	22	20 - 24	22	0.5 - 57
$ZnSO_4$				
≤0.1	A	–	A	–
0.2	527	459 - 694	527	459 - 694
0.5	312	294 - 334	350	300 - 408
1	177	155 - 195	145	129 - 154
5	93	67 - 125	109	94 - 122
10	< 48	100% mortality	< 48	100% mortality
DNP				
≤0.5	A	–	–	–
1	1416	999 - 2559	–	–
5	116	12 - 119	–	–
10	46	2 - 77	–	–
20	< 24	100% mortality	–	–

[1] ET_{50} - effective median response time of the larvae.

[2] Concentrations in which the larval ET_{50}'s were asymptotic to the log ET_{50} axis of the ILL graph.

obtained in the bench-test (Table 4). Thus the need for a controlled environment may not be as necessary for estimating the ILL as it is for determining the LC_{50}.

The LC_{50} and ILL estimates obtained using *A. aegypti* larvae compare favorably with those published in the literature for fish; examples of the latter are given in Tables 2 and 3. We suggest that these larvae can be used as test organisms for screening liquid effluents for their potential toxicity, thereby reducing the number of concentrations which have to be included in subsequent fish bioassays.

Guthrie and Wiewel (1977) expressed concern that the sensitivity of *A. aegypti* larvae to the terphenyl compound which they studied, may not be representative of the sensitivity of other aquatic invertebrates to this coolant. Our results, however, do suggest that the sensitivities of *A. aegypti* larvae and some fish species to $CuSO_4$, $ZnSO_4$ or DNP, are similar.

Table 4. Incipient lethal levels (ILL) for *Aedes aegypti* larvae, instars I to IV, reared in various toxicants either in a constant environment (Chamber) or on the laboratory bench (Bench). Also shown are published ILL's for juvenile salmon.

	Incipient lethal levels			
	A. aegypti	Juvenile Salmon, *Salmo salar*[1]		
Toxicant	Concentration (mg/l)	Concentration (mg/l)	Remarks	Reference
$CuSO_4$ (Chamber)	$> 0.02; < 0.05$	0.032	Soft water; 17°C	Sprague & Ramsey (1965)
$CuSO_4$ (Bench)	$> 0.02; < 0.05$	–	–	–
$ZnSO_4$ (Chamber)	$> 0.10; < 0.20$	0.420	Soft water; 17°C	Sprague & Ramsey (1965)
$ZnSO_4$ (Bench)	$> 0.10; < 0.20$	–	–	–
DNP (Chamber)	$> 0.50; < 1.00$	*	–	–

[1] Published values.

*No ILL of DNP for aquatic organisms found in the literature.

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STUDIES ON POTENTIAL TRANSOVARIAL TRANSMISSION OF WESTERN EQUINE ENCEPHALOMYELITIS VIRUS BY *CULEX TARSALIS* COQUILLETT

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ABSTRACT: Adult females of *Culex tarsalis* Coq. were infected with Western Equine Encephalomyelitis (WEE) virus by feeding upon viremic day-old chicks. The females were allowed 2 additional non-infective blood-meals, and the offspring of each oviposition cycle were raised to the adult stage. Samples of larvae, pupae and adults up to 3 weeks of age were assayed for virus. No WEE virus was found in any of the F_1 generations reared from the first, second or third ovarian cycles.

INTRODUCTION

One hypothesis for the maintenance of arboviruses in endemic areas is transovarial transmission by the vector (Reeves 1974). The adult female transmits the virus to her eggs while they are within the ovary (Fine 1975, Spielman 1975). The disease agent must be carried through each sequential developmental stage to the adult, and be present in sufficient concentration to be transmitted by bite to susceptible hosts. If this occurs, then transovarial transmission can be a significant factor in virus maintenance, especially in temperate zones where vector activity is periodically reduced by adverse climatic conditions.

Research on transovarial transmission with arboviruses indicates that this mechanism is possible for some encephalitis viruses. Laboratory studies on California Encephalitis Virus (CEV) in *Aedes* species, as well as supportive field isolations from larvae suggest that CEV can be maintained in nature by this mechanism (Beaty and Thompson 1976; LeDuc *et al.* 1975; McLean 1975). Venezuelan Equine Encephalitis (VEE) virus has been isolated from egg rafts laid by *Mansonia perturbans* (Walker) (Chamberlain *et al.* 1956). *Culex quinquefasciatus* Say has been reported to transmit St. Louis Encephalitis (SLE) virus to its offspring. None of the F_1 adults were infected; however, there was a low infection rate among the immature stages and most of the virus was found on the surface of the eggs (Chamberlain *et al.* 1964). Results on transovarial transmission of Western Equine Encephalomyelitis (WEE) virus by *Aedes aegypti* (L.) (Merrill and Ten Broeck 1935) and *C. tarsalis* Coq. (Barnett 1956; Chamberlain and Sudia 1957; Thomas 1963) have been negative. However, eggs laid by *A. triseriatus* (Say) infected with WEE were positive for virus (Kissling *et al.* 1957).

C. tarsalis is considered the principal epidemic vector of WEE in many parts of western North America (Chamberlain and Sudia 1957; Reeves and Hammon 1962). The importance of *C. tarsalis* in the epidemiology of WEE would be amplified if it was capable of transmitting the virus transovarially. The purpose of the following study was to determine whether a Manitoba strain of *C. tarsalis* could transmit a local isolate of WEE to its offspring.

MATERIALS AND METHODS

Virus

The WEE strain used in the 3 experiments was originally isolated from a horse brain during the epidemic of WEE in Manitoba in 1975. The strain had undergone 3 successive

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passages in Vero cell (green monkey kidney) tissue culture. Identity of the virus was confirmed by the National Arbovirus Reference Centre, Toronto, Ontario, Canada. The titre of the stock virus was $\log_{10}$ 5.3 $TCID_{50}$ (Tissue Culture Infective Dose) per 0.2 ml.

Cell Culture

Virus detection in mosquitoes and titrations of donor chick blood samples were done on Vero cells which were originally obtained from the American Type Culture Collection. The growth medium was #1969 containing 10% bovine serum, 1% L-glutamine and antibiotics: 20,000 ug of penicillin, 10,000 ug of streptomycin, 0.5 ug of neomycin and 5,000 IU of mycostatin per 100 ml. It was buffered with 8% sodium bicarbonate and 10% HEPES (N-2-Hydroxyethylpiperazine-N^1-2-ethanesulfonic acid) solution.

The cells were routinely transferred by trypsinization (Lennette and Schmidt 1969) with a 0.25% trypsin solution. Each plastic flask yielded a maximum of 50 tubes of Vero cells, each with 2 ml of media cell suspension, for virus assay the following day. Infected cells were maintained in a medium similar to the growth medium, but with 1% bovine serum and no L-glutamine (maintenance medium).

Chicks

The day-old chicks used as a source of virus for the female mosquitoes were obtained from a local hatchery. They were inoculated intra-muscularly with 0.03 ml tissue culture fluid containing 1000 $TCID_{50}$ of virus, 18 hours before the mosquitoes were allowed to feed on them. One chick was sacrificed at the beginning of the feeding period, and the rest were killed at the end, a total of 9 hours apart. The chick blood was diluted 1:4 in the anticoagulant Alsevers (Lennette and Schmidt 1969) with double strength antibiotics. These samples were frozen at -70°C, before they were centrifuged at 4000 rpm, and then titred on Vero cells.

Mosquitoes

The colony of *C. tarsalis* used in the experiments was from a colony originating in Manitoba and kept at the Department of Entomology, University of Manitoba. All larvae were reared under a photoperiod of L:D 16:8. The adults were maintained at the same photoperiod and at 75% relative humidity.

All female mosquitoes that had fed upon each group of infected chicks were caged separately. Styrofoam cups, filled with water, were provided for oviposition for a period of 48 hours beginning 4 days after each blood-meal. The female mosquitoes were offered two additional non-infective blood-meals following the first infective one.

The egg rafts were collected after each oviposition cycle, and the larvae were reared to different developmental stages before virus assay. In experiment 1, all larvae were reared to adult, and were 1 week old when frozen at -70°C. During experiments 2 and 3, larvae, in all instars, and pupae were sampled and tested for virus. Finally, adults that ranged in age from 1 to 3 weeks were tested for virus.

Viral Assay

All mosquitoes were crushed with an electric stirrer, in small (bijou) bottles containing small plastic beads and diluent. The diluent was a medium known as MEM containing 20% bovine serum and double strength antibiotics.

The infected females were ground in 1 ml of diluent after the third ovarian cycle. All offspring were ground in 2 ml of diluent after pooling (not exceeding 54 individuals). The adults were sexed before pooling. The pooled extracts were centrifuged at 4000 rpm for 20 min. before the supernatant was inoculated onto Vero cell tissue culture.

Blood samples from chicks were titred by inoculation of 10-fold dilutions onto Vero cell tubes.

All inocula were allowed to absorb for at least 1/2 h at room temperature. The cells were rinsed with Hanks medium containing double strength antibiotics and maintenance medium was finally added. The inoculated monolayers of Vero cells were observed for cytopathic effects for 6-7 days. The $TCID_{50}$ endpoints were determined by the Kärber method (Lennette and Schmidt 1969). The identity of virus recovered from the host blood samples and the mosquitoes was confirmed by the neutralization test with chick antisera on Vero cells (Lennette and Schmidt 1969).

RESULTS AND DISCUSSION

In experiments 1 and 2, all the parent female mosquitoes surviving the third ovarian cycle were positive for virus. Only 50% of those in experiment 3 contained virus; this low percentage positive was probably due to low viremia in the donor chicks which ranged from 0 to $\log_{10}$ 5.2 $TCID_{50}$. Viremias in the other donor chicks were $\log_{10}$ 5.1-7.1 $TCID_{50}$ for experiment 1, and $\log_{10}$ 6.2-7.5 $TCID_{50}$ for experiment 2.

There was no evidence of transovarial transmission in any of the 3 experiments. All pools of larvae, pupae, and adults were negative (Table 1). The lack of virus in the F_1 generations of infected females in these experiments agrees with research on WEE in the United States with *C. tarsalis* (Chamberlain and Sudia 1957; Reeves and Hammon 1962; Thomas 1963).

Thomas (1963) found that after *C. tarsalis* females took an infective blood-meal, no WEE virus was detected in the ovaries until 4 days later. Virus concentration reached a maximum 10 days after the blood-meal. Theoretically then, progeny arising from eggs laid at least 10 days following the infective blood-meal would be more likely to be infected than those resulting from eggs laid earlier. In *C. quinquefasciatus* for example, Chamberlain *et al.* (1964) found that SLE virus was absent on eggs laid before the 8th day following adult infection but 92% of the eggs laid after 8 days were contaminated with virus. Most of the virus was found to be on the outside of the eggs. It is interesting that Thomas (1963) found a few *C. tarsalis* egg rafts, laid by WEE infected adults, to harbour the virus. Virus was present in the ovaries of infected adults and this may have resulted in surface contamination of the eggs. This would explain why no virus was found in the immature or adult offspring in any of his trials.

There remains the possibility that hatched first-instar larvae could become infected by feeding on the surface of egg rafts contaminated with virus. Mosquito larvae in the laboratory have been infected by placing them in media containing virus (Collins 1963; Peleg 1965). Adult females reared from these larvae were able to transmit the virus to a host.

Table 1. Number of mosquito pools and mosquitoes tested for transovarial transmission of Western Equine Encephalomyelitis.

	Ovarian cycles								
Developmental	Exp. 1.			Exp. 2.			Exp. 3.		
stage	1	2	3	1	2	3	1	2	3
Larvae				3/68	6/162	4/103	3/57	6/143	3/71
Pupae				6/144	1/28	2/52	2/43	5/119	1/28
Adults									
1 wk old	5[1]/230	13/607	8/373	5/214	3/70	2/27	4/100	2/44	2/53
2 wk old				3/109	2/40	2/22	4/79	2/29	4/75
3 wk old				3/94	2/51		2/29	2/20	

[1] # of pools/total # of mosquitoes tested.

Most of the evidence available to date suggests that transovarial transmission of WEE virus by *C. tarsalis* does not occur. No WEE virus has been isolated from male *C. tarsalis*, caught in the wild, although it is known that they harbour the virus for at least 1 week (Reeves and Hammon 1962). Also, no virus has been isolated from female adults raised from field-collected *C. tarsalis* larvae (Hammon and Reeves 1947). Lastly, Watts and Eldridge (1975) have suggested that there is greater selection pressure for a virus to be transmitted transovarially to the egg in mosquitoes that overwinter in the egg stage than in mosquitoes like *C. tarsalis* that overwinter in the adult stage. Beaty and Thompson (1976) have shown that the La Crosse strain of CEV is indeed transmitted transovarially to the eggs of *A. triseriatus,* a species that overwinters in the egg stage. If *C. tarsalis* females were to be involved in overwintering WEE virus, they could pass on the virus to new hosts in spring through an infective bite. Transovarial transmission would therefore be of little or no selective advantage in the maintenance of a virus in mosquitoes that overwinter as adults.

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ANT-APHID ASSOCIATIONS IN THE PROVINCE OF MANITOBA

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ABSTRACT: Twenty-seven species of ants tending 62 species of aphids were recorded from Manitoba. Ants of the genus *Formica* were the dominant aphid attendants. Aphids of the genus *Aphis* were the most frequently attended. Nearly 10% of the associations consisted of two different species or genera of ants together; usually one was the true aphid attendant and the other a scavenger species.

INTRODUCTION

Few surveys have been conducted to determine North American ant-aphid associations (Neilsson *et al.* 1971). Jones (1929) reported that in Colorado 149 species of aphids in 34 genera were tended in different combinations by 92 species of ants in 15 genera. As in Europe (Atanassov 1974, Egger 1973, Fossel 1972, Horstmann 1972, Kloft 1959) and the Middle East (Bodenheimer and Swirski 1957), ants are associated with most Canadian aphid species of the genus *Cinara* (LACHNIDAE) (Bradley 1961, McNeil *et al.* 1977). Bodenheimer and Swirski (1957) stated that aphids of the genera *Pemphigus, Prociphilus* and *Thecabius* (PEMPHIGIDAE) are not tended by ants in the Middle East; however, one species of *Prociphilus* is tended by the ant *Acanthomyopes latipes* (Walsh) in Manitoba (Bradley and Hinks 1968), and *Prociphilus* (=*Paraprociphilus*) *tesselatus* (Fitch) is tended by three species of ants in New York State (Eisner *et al.* 1978). Gregg (1972) reported that 43 species and subspecies of ants were present in the Canadian zone and many of these are known aphid attendants. Ant-aphid associations of Manitoba, found as a result of this study, are enumerated and discussed.

METHODS

Areas throughout Manitoba were examined for ant-aphid associations during 1976 and 1977. The aerial portions of plants were inspected for aphid colonies being tended by ants. A small portion of the plant bearing the ants and aphids was placed in a numbered polyethylene bag, taken to the laboratory and stored at 0°C. The aphids and ants were later transferred to vials containing 70% ethyl alcohol. Aphids were processed for identification following the method outlined by Richards (1964) and were named in accordance with Eastop and Hille Ris Lambers (1976).

A. Francoeur (personal communication) stated that all material from Manitoba identified as *Formica fusca* L. should be named *F. subaenescens* Emery.

RESULTS AND DISCUSSION

Identification of the collections indicated that 241 different ant-aphid associations were found (Tables 1 and 4). Twenty-seven species in 7 genera of ants collected honeydew from 62 species in 28 genera of aphids on 39 host plants within the province of Manitoba (Table 1).

Formicidae

As reported by Jones (1929) for Colorado, ants of the genus *Formica* were the dominant and least species-specific aphid attendants in Manitoba. Fourteen species of *Formica* collected honeydew from 55 species of aphids. The second most prevalent attendants belonged to 4 species of *Lasius,* found tending 22 species of aphids; these were followed by 3 species of *Camponotus,* collected with 17 species of aphids. Two species each of *Myrmica* and *Dolichoderus* tended 19 and 11 species of aphids, respectively.

Table 1. Species of aphids, attendant ants, host plants, and locations and number of ant-aphid associations collected in Manitoba in 1976 and 1977.

Aphids and ants	Host plants	Number of records
Aphis sp.	*Spiraea alba*[1]; *Artemisia* sp.[2]	
[1] *Dolichoderus taschenbergi*	Sandilands Prov. Forest	1
[2] *Lasius neoniger*	" " "	1
Aphis armoraciae Cowen	*Rudbeckia hirta*[1]; *Picea glauca*[2]	
[2] *Formica obscuripes*	Sandilands Prov. Forest	1
[1] *Formica oreas comptula*	Carberry	1
Aphis asclepiadis Fitch	*Apocynum cannabinum*	
Myrmica brevispinosa	Birds Hill Prov. Park	1
Formica podzolica	Sandilands Prov. Forest	1
Aphis citricola van der Goot	*Spiraea* sp.	
Lasius pallitarsis	Winnipeg (City)	1
Aphis fabae Scopoli	*Viburnum opulus nanum*[1]; *Philadelphus coronarius; Arctium minus*[3]	
[3] *Myrmica brevispinosa*	Morden	1
[1,2] *Lasius pallitarsis*	Winnipeg (UM Campus; City)	3
[2,3] *Formica podzolica*	Winnipeg (City); Morden	2
Aphis farinosa Gmelin	*Populus balsamifera*[1]; *Salix* sp.[2]; *Salix planifolia*[3]	
[2] *Lasius pallitarsis*	Spruce Woods Prov. Forest	1
[1] *Formica podzolica*	Sandilands Prov. Forest	1
[3] *Formica neorufibarbis*	Churchill	2
Aphis gossypii Glover	*Galium boreale*[1]; *Oenothera biennis*[2]; *Diervilla lonicera*[3]	
[1,3] *Myrmica emeryana*	Whiteshell Prov. Park (Hanson Creek)	3
[3] *Dolichoderus plagiatus*	Pinawa (Atomic Energy FIG Area)	2
[3] *Tapinoma sessile*	Agassiz Prov. Forest	1
[3] *Camponotus noveboracensis*	" " "	1
[2] *Lasius alienus*	Pinawa (Atomic Energy FIG Area)	1
[2] *Formica lasioides*	" " " " "	1
[3] *Formica subnuda*	" " " " "	1
[2] *Formica obscuripes*	Sandilands Prov. Forest	1
[1] *Formica (rufa)* species?	" " "	2
[2] *Formica subsericea*	Whitemouth Lake	1
[3] *Formica subaenescens*	Pinawa (Atomic Energy FIG Area)	1
Aphis helianthi Monell	*Cornus stolonifera*	
Camponotus nearcticus	Sandilands Prov. Forest	1
Lasius pallitarsis	Winnipeg (UM Campus)	1
Formica podzolica	Birds Hill Prov. Park	1
Formica subaenescens	" " " "	1
Aphis knowltoni Hottes and Frison	*Taraxacum officinale*	
Myrmica brevispinosa	Beaconia	1
Aphis maculatae Oestlund	*Populus tremuloides*[1]; *P. balsamifera*[2]; *Populus* sp.[3]	
[1] *Myrmica emeryana*	Sandilands Prov. Forest; Whiteshell Prov. Park (Hanson Creek)	2
[3] *Tapinoma sessile*	Whiteshell Prov. Park (Lone Island Lake)	1
[1] *Camponotus herculeanus*	Northwest Angle Prov. Park	1

continued next page

Table 1. (continued)

Aphids and ants	Host plants	Number of records
A. maculatae (continued)		
[1,2,3] *Camponotus noveboracensis*	Camp Morton; Rennie; Winnipeg (City) Sandilands Prov. Forest; LaBarriere Park; Whiteshell Prov. Park (Lone Island Lake)	7
[2] *Lasius pallitarsis*	Sandilands Prov. Forest	1
[1] *Formica lasioides*	" " "	1
[1] *Formica subnuda*	Whiteshell Prov. Park (Lone Island Lake)	1
[1] *Formica (rufa)* species?	Sandilands Prov. Forest	1
[1] *Formica subsericea*	Whitemouth Lake	1
[1,3] *Formica subaenescens*	Northwest Angle Prov. Park; Whiteshell Prov. Park (Lone Island Lake); Sandilands Prov. Forest	10
Aphis neogillettei Palmer	*Cornus stolonifera*	
Myrmica emeryana	Spruce Woods Prov. Forest	1
Dolichoderus plagiatus	Birds Hill Prov. Park	1
Tapinoma sessile	Richer; Sandilands Prov. Forest	2
Camponotus herculeanus	Rennie	1
Camponotus noveboracensis	Birds Hill Prov. Park; Sandilands Prov. Forest; Hnausa Park	3
Lasius alienus	Rennie; Birds Hill	2
Lasius pallitarsis	Winnipeg (UM Campus); Spruce Woods Prov. Forest; Stoney Mountain; Whiteshell Prov. Park (Hanson Creek)	4
Lasius subumbratus	Spruce Woods Prov. Forest	1
Formica lasioides	Rennie	1
Formica subnuda	Richer; Hecla Island	2
Formica obscuripes	Morden; Stoney Mountain	2
Formica oreas comptula	Spruce Woods Prov. Forest; Birds Hill Prov. Park; Birds Hill; Aweme	5
Formica podzolica	Birds Hill; Richer; Stoney Mountain; Birds Hill Prov. Park; Winnipeg (UM Campus)	5
Formica subaenescens	Aweme	1
Aphis oenotherae Oestlund	*Epilobium angustifolium*	
Formica podzolica	Thompson	2
Aphis oestlundi Gillette	*Oenothera biennis*	
Formica obscuripes	Sandilands Prov. Park	1
Formica subsericea	" " "	1
Aphis rubicola Oestlund	*Rubus strigosus*	
Myrmica emeryana	Birds Hill Prov. Park	1
Camponotus noveboracensis	Sandilands Prov. Forest; Birds Hill Prov. Park	3
Formica podzolica	Sandilands Prov. Forest	1
Aphis spiraephila Patch	*Spiraea alba*	
Myrmica emeryana	Whiteshell Prov. Park (Hanson Creek)	1
Lasius alienus	" " " " "	1
Lasius pallitarsis	" " " " "	1
Formica podzolica	" " " " " Sandilands Prov. Forest	3

continued next page

Table 1. (continued)

Aphids and ants	Host plants	Number of records
Aphis varians Patch	*Epilobium angustifolium*	
Myrmica emeryana	Whiteshell Prov. Park (Hanson Creek)	1
Camponotus noveboracensis	Pinawa (Atomic Energy FIG Area); Northwest Angle Prov. Park; Whitemouth Lake	5
Formica lasioides	Whitemouth Lake	1
Formica obscuripes	Sandilands Prov. Forest	1
Formica oreas comptula	Agassiz Prov. Forest	1
Formica podzolica	Pinawa (Atomic Energy FIG Area); Whiteshell Prov. Park (Hanson Creek)	4
Formica subsericea	Whitemouth Lake	2
Formica subaenescens	Northwest Angle Prov. Park	3
Formica neorufibarbis	Churchill	1
Aphis viburniphila Patch	*Viburnum rafinesquianum*[1]; *V. trilobum*[2]	
[1]*Formica obscuripes*	Morden	2
[1]*Formica oreas comptula*	Morden; Birds Hill Prov. Park	2
[2]*Formica montana*	Winnipeg (UM Campus)	1
Aphis whiteshellensis Rojanavongse and Robinson	*Amelanchier alnifolia*	
Myrmica emeryana	Whiteshell Prov. Park (Hanson Creek)	2
Formica subaenescens	" " " " "	1
Aphthargelia symphoricarpi (Thomas)	*Symphoricarpos occidentalis*	
Camponotus noveboracensis	Stoney Mountain	1
Lasius alienus	Morden	4
Lasius pallitarsis	Spruce Woods Prov. Forest	2
Formica lasioides	Sandilands Prov. Forest	1
Formica obscuripes	" " "	1
Formica ulkei	Cook's Creek	1
Formica podzolica	Stoney Mountain	1
Asiphonaphis pruni Wilson and Davis	*Prunus virginiana*[1]; *P. pensylvanica*[2]	
[1]*Lasius alienus*	Carberry	1
[1,2]*Formica obscuripes*	Sandilands Prov. Forest; Morden	5
[1]*Formica oreas comptula*	" " "	1
[1]*Formica (rufa)* species?	" " "	1
Asiphum tremulae (L.)	*Populus tremuloides*	
Dolichoderus taschenbergi	Sandilands Prov. Forest	1
Formica subnuda	" " "	1
Formica oreas comptula	" " "	1
Formica podzolica	" " " ; Thompson	3
Formica subaenescens	" " "	1
Ceruraphis viburnicola Gillette	*Viburnum opulus nanum*	
Formica podzolica	Winnipeg (UM Campus)	1
Chaitophorus sp. *(pustulatus* grp.*)*	*Salix* sp.	
Formica oreas comptula	Aweme	1
Chaitophorus macrostachyae (Essig)	*Salix* sp.	
Formica obscuriventris	Morden	2

continued next page

Table 1. (continued)

Aphids and ants	Host plants	Number of records
Chaitophorus neglectus Hottes and Frison	*Populus tremuloides*	
Formica subnuda	Sandilands Prov. Forest	1
Formica subaenescens	" " "	1
Chaitophorus nigrae Oestlund	*Salix* sp.[1]; *Salix bebbiana*[2]	
[1] *Myrmica brevispinosa*	Birds Hill Prov. Park	2
[1] *Dolichoderus plagiatus*	" " " "	1
[1] *Camponotus noveboracensis*	Hecla Island	1
[1] *Lasius alienus*	Whiteshell Prov. Park (Hanson Creek)	1
[1] *Lasius pallitarsis*	Spruce Woods Prov. Forest	1
[1] *Formica subnuda*	" " " "	1
[1] *Formica obscuripes*	Lewis	3
[2] *Formica oreas comptula*	Birds Hill Prov. Park	12
[1] *Formica (rufa)* species?	Sandilands Prov. Forest	1
[1] *Formica hewitti*	Northwest Angle Prov. Park	1
[1] *Formica podzolica*	Thompson; Birds Hill Prov. Park; Sandilands Prov. Forest; Birds Hill	4
[1] *Formica subaenescens*	Richer	1
Chaitophorus nigricentrus Richards	*Salix* sp.[1]; *Salix bebbiana*[2]	
[1] *Formica subnuda*	Birds Hill Prov. Park	1
[1] *Formica obscuripes*	Sandilands Prov. Forest	1
[1] *Formica oreas comptula*	Birds Hill Prov. Park; Birds Hill	11
[1] *Formica podzolica*	Birds Hill	1
[1] *Formica subaenescens*	Birds Hill Prov. Park	1
Chaitophorus nudus Richards	*Populus tremuloides*	
Lasius alienus	Carberry	1
Formica oreas comptula	Sandilands Prov. Forest; Agassiz Prov. Forest; Carberry; Aweme; Birds Hill Prov. Park	31
Formica (rufa) species?	Carberry; Sandilands Prov. Forest	2
Chaitophorus populicola Thomas	*Populus tremuloides*[1]; *P. balsamifera*[2]; *Populus* sp.[3]	
[2,3] *Myrmica brevispinosa*	Portage La Prairie	5
[1] *Myrmica emeryana*	Sandilands Prov. Forest; Whiteshell Prov. Park (Hanson Creek); Birds Hill Prov. Park	5
[1] *Leptothorax muscorum*	Whiteshell Prov. Park (Hanson Creek)	1
[2] *Dolichoderus taschenbergi*	Aweme	1
[1,2] *Tapinoma sessile*	Agassiz Prov. Forest; Sandilands Prov. Forest	2
[1] *Camponotus herculeanus*	Northwest Angle Prov. Park	1
[1,2,3] *Camponotus noveboracensis*	Agassiz Prov. Forest; Aweme; Camp Morton; Carberry; Fortier; LaBarriere Park; Lewis; Winnipeg (City); Pinawa (Atomic Energy FIG Area); Portage La Prairie; Rennie; Sandilands Prov. Forest; Stoney Mountain; Whitemouth Lake; Whiteshell Prov. Park (Hanson Creek; Lone Island Lake)	25

continued next page

Table 1. (continued)

Aphids and ants	Host plants	Number of records
C. populicola (continued)		
[1,2] *Lasius alienus*	Pinawa (Atomic Energy FIG Area); Portage La Prairie; Sandilands Prov. Forest; Whiteshell Prov. Park (Hanson Creek)	6
[1,2] *Formica lasioides*	Sandilands Prov. Forest; Rathwell	3
[1,2] *Formica subnuda*	Fortier; Hecla Island; Sandilands Prov. Forest; Northwest Angle Prov. Park; Whiteshell Prov. Park (Hanson Creek); Birds Hill Prov. Park	9
[2,1] *Formica obscuripes*	Sandilands Prov. Forest; Portage La Prairie; Stoney Mountain; Lewis	8
[3] *Formica obscuriventris*	Morden	1
[2,1] *Formica oreas comptula*	Carberry; Aweme; Birds Hill Prov. Park; Birds Hill	30
[1] *Formica (rufa)* species?	Sandilands Prov. Forest	2
[1] *Formica hewitti*	Hecla Island	1
[1,2] *Formica podzolica*	Birds Hill; Sandilands Prov. Forest; Northwest Angle Prov. Park	5
[1,2] *Formica subsericea*	Whitemouth Lake; Lewis	4
[1,3] *Formica subaenescens*	Whiteshell Prov. Park (Lone Island Lake); Sandilands Prov. Forest; Northwest Angle Prov. Park	11
Chaitophorus populifolii (Essig)	*Populus tremuloides*[1]; *P. balsamifera*[2]; *Salix bebbiana*[3]	
[1] *Myrmica emeryana*	Sandilands Prov. Forest; Whiteshell Prov. Park (Hanson Creek)	2
[1] *Camponotus noveboracensis*	LaBarriere Park; Stoney Mountain; Dacotah	3
[1] *Lasius alienus*	Sandilands Prov. Forest	1
[1] *Formica obscuripes*	Stoney Mountain	1
[1,2,3] *Formica oreas comptula*	Birds Hill Prov. Park	4
[1] *Formica podzolica*	Northwest Angle Prov. Park	1
Chaitophorus saliciniger (Knowlton)	*Cornus stolonifera*[1]; *Salix* sp.[2]; *Salix bebbiana*[3]	
[3] *Formica oreas comptula*	Birds Hill Prov. Park	20
[2,1] *Formica podzolica*	Sandilands Prov. Forest; Thompson; Birds Hill Prov. Park	6
[3] *Formica hewitti*	" " " "	1
Cinara sp.	*Pinus banksiana; Picea glauca*	
Formica oreas comptula	Sandilands Prov. Forest; Birds Hill Prov. Park	2
Cinara banksiana Pepper and Tissot	*Pinus banksiana*	
Dolichoderus taschenbergi	Sandilands Prov. Forest	1
Lasius alienus	" " "	1
Formica obscuripes	" " "	5
Formica podzolica	" " "	1
Cinara braggi (Gillette)	*Picea glauca*	
Formica podzolica	Carberry	1

continued next page

Table 1. (continued)

Aphids and ants	Host plants	Number of records
Cinara canatra Hottes and Bradley	*Pinus banksiana*	
Formica obscuripes	Sandilands Prov. Forest	2
Formica oreas comptula	" " " ; Agassiz Prov. Forest	2
Cinara coloradensis (Gillette)	*Picea glauca*	
Dolichoderus taschenbergi	Carberry	1
Cinara fornacula Hottes	*Picea glauca*	
Formica lasioides	Sandilands Prov. Forest	1
Formica obscuripes	" " "	1
Formica podzolica	Whiteshell Prov. Park (Hanson Creek) Northwest Angle Prov. Park; Sandilands Prov. Forest	5
Formica subaenescens	Northwest Angle Prov. Park	1
Cinara laricifex (Fitch)	*Larix laricina*	
Camponotus herculeanus	Churchill	5
Formica densiventris	Sandilands Prov. Forest	1
Formica subnuda	" " "	1
Formica podzolica	" " "	4
Formica hewitti	" " "	1
Formica neorufibarbis	Churchill	3
Cinara obscura Bradley	*Picea glauca*	
Formica obscuripes	Lewis	1
Formica neorufibarbis	Churchill	1
Cinara pergandei (Wilson)	*Pinus banksiana*	
Camponotus noveboracensis	Lewis; Sandilands Prov. Forest	4
Formica subnuda	Pinawa (Atomica Energy FIG Area)	1
Formica obscuripes	Sandilands Prov. Forest	1
Formica obscuriventris	Agassiz Prov. Forest	1
Formica oreas comptula	" " " ; Whitemouth Lake	2
Formica podzolica	Pinawa (Atomic Energy FIG Area)	1
Cinara petersoni Bradley	*Juniperus horizontalis*	
Formica subnuda	Carberry	1
Formica spatulata	Rathwell	1
Cinara spiculosa Bradley	*Larix laricina*	
Camponotus herculeanus	Churchill	1
Formica neorufibarbis	"	1
Hamamelistes spinosus Shimer	*Betula glandulosa* var. *glandulifera*	
Formica oreas comptula	Birds Hill Prov. Park	1
Hoplochaitophorus quercicola (Monell)	*Quercus macrocarpa*	
Myrmica brevispinosa	Morden	1
Camponotus noveboracensis	" ; Spruce Woods Prov. Forest	2
Lasius alienus	"	2
Formica obscuripes	Carberry	2
Formica oreas comptula	Whitemouth Lake; Birds Hill Prov. Park; Aweme	5
Formica podzolica	Morden	1
Formica subsericea	Whitemouth Lake	2

continued next page

Table 1. (continued)

Aphids and ants	Host plants	Number of records
Hysteroneura setariae (Thomas)	*Prunus*[1]; *Prunus pumila*[2]	
[2] *Tapinoma sessile*	Sandilands Prov. Forest	1
[1] *Lasius pallitarsis*	Winnipeg (City)	1
[2] *Formica podzolica*	Sandilands Prov. Forest	1
Lachnus allegheniensis McCook	*Quercus macrocarpa*	
Formica subnuda	Birds Hill	1
Formica podzolica	" "	1
Macrosiphoniella sp.	*Artemisia frigida*	
Formica subsericea	Whitemouth Lake	1
Macrosiphoniella absinthii (L.)	*Artemisia* sp.	
Formica lasioides	Stoney Mountain	1
Maculolachnus sijpkensi Hille Ris Lambers	*Rosa acicularis*	
Formica obscuripes	Sandilands Prov. Forest	4
Formica oreas comptula	Birds Hill Prov. Park; Birds Hill	3
Mastopoda pteridis Oestlund	*Pteridium aquilinum*	
Myrmica emeryana	Birds Hill Prov. Park	1
Meliarhizophagus fraxinifolii (Riley)	*Fraxinus* sp.	
Camponotus noveboracensis	Morden	1
Myzocallis punctatus (Monell)	*Quercus macrocarpa*	
Formica oreas comptula	Sandilands Prov. Forest	1
Formica subsericea	Whitemouth Lake	1
Myzus cerasi (Fabricius)	*Prunus pensylvanica*	
Dolichoderus taschenbergi	Carberry	1
Dolichoderus plagiatus	Rathwell	1
Tapinoma sessile	Sandilands Prov. Forest	1
Camponotus herculeanus	Pinawa (Atomic Energy FIG Area)	2
Camponotus noveboracensis	Sandilands Prov. Forest	1
Lasius alienus	Whiteshell Prov. Park (Hanson Creek); Birds Hill	2
Formica lasioides	Sandilands Prov. Forest	1
Formica subnuda	Rennie; Northwest Angle Prov. Park	2
Formica obscuripes	Sandilands Prov. Forest	2
Formica oreas comptula	Whitemouth Lake	2
Formica podzolica	Sandilands Prov. Forest; Pinawa (Atomic Energy FIG Area); Rennie; Carberry	6
Formica subaenescens	Agassiz Prov. Forest	1
Formica subsericea	Whitemouth Lake	1
Nearctaphis sp.	*Crataegus* sp.	
Formica oreas comptula	Birds Hill Prov. Park	1
Nearctaphis clydesmithi Hille Ris Lambers	*Crataegus* sp.	
Formica obscuripes	Stoney Mountain	1
Nearctaphis crataegifoliae (Fitch)	*Crataegus* sp.	
Formica obscuripes	Sandilands Prov. Forest	3

continued next page

Table 1. (continued)

Aphids and ants	Host plants	Number of records
Nearctaphis sensoriata (Gillette and Bragg)	*Amelanchier alnifolia*	
Lasius alienus	Agassiz Prov. Forest	1
Formica oreas comptula	Sandilands Prov. Forest	1
Neosymydobius mimicus Hottes	*Quercus macrocarpa*	
Dolichoderus taschenbergi	Carberry	1
Paraprociphilus tesselatus (Fitch)	*Alnus rugosa*	
Formica podzolica	Sandilands Prov. Forest	1
Pemphigus sp.	*Populus tremuloides*	
Dolichoderus taschenbergi	Sandilands Prov. Forest	1
Periphyllus negundinis (Thomas)	*Acer negundo*	
Formica subnuda	Fortier	1
Prociphilus sp.	Moss	
Myrmica emeryana	Sandilands Prov. Forest	1
Prociphilus erigeronensis (Thomas)	?Grass roots permeating ant nest	
Lasius pallitarsis	Fortier	1
Pseudopterocomma canadensis Richards	*Populus tremuloides*	
Dolichoderus taschenbergi	Sandilands Prov. Forest	1
Pterocomma bicolor (Oestlund)	*Populus balsamifera*[1]; *Salix* sp.[2]; *Populus* sp.[3]	
[3] *Myrmica brevispinosa*	Portage La Prairie	1
[1] *Camponotus noveboracensis*	Sandilands Prov. Forest; Birds Hill Prov. Park; Whiteshell Prov. Park (Hanson Creek)	6
[2] *Lasius subumbratus*	Spruce Woods Prov. Forest	1
[1] *Formica subnuda*	Sandilands Prov. Forest	1
[1] *Formica oreas comptula*	Birds Hill Prov. Park	2
[1] *Formica subaenescens*	" " " "	2
Pterocomma smithiae (Monell)	*Populus balsamifera*[1]; *Salix* sp.[2]; *Salix planifolia*[3]	
[2] *Lasius pallitarsis*	Spruce Woods Prov. Forest	1
[1] *Formica oreas comptula*	Birds Hill Prov. Park	29
[3] *Formica neorufibarbis*	Churchill	1
Rhopalosiphum sp.	*Crataegus* sp.	
Formica oreas comptula	Birds Hill Prov. Park	1
Rhopalosiphum cerasifoliae (Fitch)	*Prunus virginiana*	
Myrmica emeryana	Cook's Creek	1
Formica subnuda	Sandilands Prov. Forest	1
Formica oreas comptula	" " "	1

continued next page

Table 1. (continued)

Aphids and ants	Host plants	Number of records
Symydobius americanus Baker	*Betula glandulosa* var. *glandulifera*[1]; *B. papyrifera*[2]	
[1] *Myrmica emeryana*	Birds Hill Prov. Park	1
[1] *Tapinoma sessile*	" " " "	1
[2] *Formica obscuripes*	Lewis	2
[1] *Formica oreas comptula*	Birds Hill Prov. Park	21
Formica hewitti and/or *F. podzolica*	" " " "	*
Thecabius affinis (Kaltenbach)	*Populus tremuloides*	
Formica podzolica	Thompson	1

[1,2,3] Host plants on which the respective species of ants were collected.

* Several visual records but no samples collected.

Certain species of *Formica* were collected more frequently and with a greater number of different aphid species than were other *Formica* species. This was also true for some species of the genera *Lasius* and *Camponotus*. For example, *Formica podzolica* Francoeur, *F. oreas comptula* Wheeler and *F. obscuripes* Forel tended 30, 25 and 23 aphid species, respectively, *F. subnuda* Emery and *F. subaenescens* tended 16 and 14 species, respectively, while *F. obscuriventris* Mayr, *F. hewitti* Wheeler and *F. subsericea* Say, respectively, tended only 3, 4 and 5 species of aphids. These differences may be a function of the distribution and/or abundance of species of ants within the province. It is also possible that the less frequently collected species of ants are more specific (less opportunistic) in the species of aphids from which they collect their honeydew. The species of ants and their collection sites are summarized in Table 2.

Table 2. Collection sites for species of ants found tending aphids in the Province of Manitoba during 1976 and 1977.

Myrmica brevispinosa Emery
Beaconia; Birds Hill Prov. Park; Lake Minnewasta; Portage La Prairie.

Myrmica emeryana Forel
Birds Hill; Birds Hill Prov. Park; Cook's Creek; Sandilands Prov. Forest; Spruce Woods Prov. Forest (Oxbow Lake Nature Trail); Whiteshell Prov. Park (Hanson Creek, Caddy Lake).

Leptothorax muscorum Provancher
Whiteshell Prov. Park (Hanson Creek).

Dolichoderus plagiatus (Mayr)
Birds Hill Prov. Park; Pinawa (Atomic Energy FIG Area); Rathwell.

Dolichoderus taschenbergi (Mayr)
Aweme; Carberry; Sandilands Prov. Forest.

Tapinoma sessile (Say)
Agassiz Prov. Forest; Richer; Sandilands Prov. Forest; Whiteshell Prov. Park (Lone Island Lake).

continued next page

Table 2. (continued)

Camponotus herculeanus (L.)
Churchill; Pinawa (Atomic Energy FIG Area); Rennie (Bird Sanctuary).

Camponotus nearcticus Emery
Sandilands Prov. Forest.

Camponotus noveboracensis (Fitch)
Agassiz Prov. Forest; Birds Hill Park; Camp Morton; Carberry; Dacotah; Fortier; Hecla Island; Hnausa Park; LaBarriere Park; Lake Minnewasta; Lewis; Little Mountain Park; Northwest Angle Prov. Park (Moose Lake); Pinawa (Atomic Energy FIG Area); Rennie (Bird Sanctuary); Sandilands Prov. Forest; Spruce Woods Prov. Forest (Oxbow Lake Nature Trail); Stoney Mountain; Whitemouth Lake; Whiteshell Prov. Park (Hanson Creek, Caddy Lake and Lone Island Lake).

Lasius alienus Forster
Agassiz Prov. Forest; Birds Hill; Carberry; Lake Minnewasta; Pinawa (Atomic Energy FIG Area); Portage La Prairie; Rennie (Bird Sanctuary); Sandilands Prov. Forest; Whiteshell Prov. Park (Hanson Creek, Caddy Lake).

Lasius pallitarsis (Provancher)
Fortier; Sandilands Prov. Forest; Spruce Woods Prov. Forest (Oxbow Lake Nature Trail); Stoney Mountain; Whiteshell Prov. Park (Hanson Creek, Caddy Lake); Winnipeg (City; UM Campus).

Lasius neoniger Emery
Sandilands Prov. Forest.

Lasius subumbratus Viereck
Spruce Woods Prov. Forest (Oxbow Lake Nature Trail).

Formica lasioides Emery
Pinawa (Atomic Energy FIG Area); Rathwell; Rennie (Bird Sanctuary); Sandilands Prov. Forest; Stoney Mountain; Whitemouth Lake.

Formica subnuda Emery
Birds Hill; Birds Hill Prov. Park; Carberry; Fortier; Hecla Island; Northwest Angle Prov. Park (Moose Lake); Pinawa (Atomic Energy FIG Area); Rennie (Bird Sanctuary); Richer; Sandilands Prov. Forest; Spruce Woods Prov. Forest (Oxbow Lake Nature Trail); Whiteshell Prov. Park (Hanson Creek, Caddy Lake and Lone Island Lake).

Formica (rufa) species?
Carberry; Sandilands Prov. Forest.

Formica obscuripes Forel
Birds Hill Prov. Park; Carberry; Lake Minnewasta; Lewis; Portage La Prairie; Sandilands Prov. Forest; Stoney Mountain.

Formica obscuriventris Mayr*
Agassiz Prov. Forest; Lake Minnewasta.

Formica oreas comptula Wheeler
Agassize Prov. Forest; Aweme; Birds Hill; Birds Hill Prov. Park; Carberry; Lake Minnewasta; Sandilands Prov. Forest; Spruce Woods Prov. Forest (Oxbow Lake Nature Trail); Whitemouth Lake.

Formica ulkei Emery
Cook's Creek.

continued next page

Table 2. (continued)

Formica densiventris Viereck
Sandilands Prov. Forest.

Formica spatulata Buren
Rathwell.

Formica podzolica Francoeur
Aweme; Birds Hill; Birds Hill Prov. Park; Carberry; Lewis; Lake Minnewasta; Northwest Angle Prov. Park (Moose Lake); Pinawa (Atomic Energy FIG Area); Rennie (Bird Sanctuary); Sandilands Prov. Forest; Stoney Mountain; Thompson; Whitemouth Lake; Whiteshell Prov. Park (Hanson Creek, Caddy Lake); Winnipeg (City; UM Campus).

Formica subaenescens Emery
Agassiz Prov. Forest; Aweme; Birds Hill Prov. Park; Northwest Angle Prov. Park (Moose Lake); Pinawa (Atomic Energy FIG Area); Richer; Sandilands Prov. Forest; Whiteshell Prov. Park (Hanson Creek, Caddy Lake and Lone Island Lake).

Formica montana Emery
Winnipeg (UM Campus).

Formica hewitti Wheeler
Birds Hill Prov. Park; Hecla Island; Northwest Angle Prov. Park (Moose Lake); Sandilands Prov. Forest.

Formica subsericea Say
Lewis; Sandilands Prov. Forest; Whitemouth Lake.

Formica neorufibarbis Emery
Churchill.

***Gregg (1972) reported the occurrence of *F. obscuriventris* in Birds Hill Prov. Park, but he did not mention the presence of *F. oreas comptula* therein. *F. obscuriventris* was not collected in Birds Hill Prov. Park in the present study, but *F. oreas comptula* was found to be well represented. Identification of the latter species was confirmed upon examination of its alate reproductives.**

In this study, 9.7% of the associations each contained two species of ants (Table 3). The collection of an association containing more than one species of ant does not necessarily imply that the ant species were interacting. Observations of the tending activities of the ants, prior to collection, revealed that one species often appeared to function as the true aphid attendant (e.g., *F. oreas comptula*) while the other species of ant was a honeydew scavenger (e.g., *F. podzolica*), foraging in the territory but actively avoiding contact with the attendant species. However, in some cases, such as with *Tapinoma sessile* (Say) with *Camponotus noveboracensis* (Fitch), the fact that more than one species of ant was involved was discovered only when the samples were later identified in the laboratory. It is possible that *Camponotus* is tolerant of the presence of certain other genera of ants, or perhaps it is *Camponotus* which is being tolerated. In one sample, 6 *F. podzolica* and 15 *C. herculeanus* (L.) were collected together. If these species were intolerant of one another, aggressive behaviour should have manifested itself in some way, either during the initial observation or during transportation to the laboratory. Several collections contained high numbers of *F. oreas comptula* and *F. subsericea,* or *F. subnuda* and *F. subaenescens.* The former species in each case is known to sometimes enslave other species of ants (Wheeler and Wheeler 1963); dulosis might therefore account for these particular mixed collections.

Table 3. Species of ants collected together while tending aphids on the same host plant.

Lasius pallitarsis	+ *Formica podzolica*
L. alienus	+ *F. lasioides*
	+ *F. obscuripes*
Camponotus noveboracensis	+ *Tapinoma sessile*
	+ *F. podzolica*
	+ *F. neorufibarbis*
	+ *F. subaenescens*
C. herculeanus	+ *F. podzolica*
	+ *F. neorufibarbis*
	+ *F. lasioides*
	+ *F. subaenescens*
F. subnuda	+ *F. podzolica*
	+ *Myrmica emeryana*
	+ *F. hewitti*
	+ *F. subaenescens*
F. oreas comptula	+ *F. podzolica*
	+ *F. subsericea*
M. brevispinosa	+ *F. podzolica*
M. emeryana	+ *T. sessile**
	+ *Leptothorax muscorum*
Dolichoderus taschenbergi	+ *F. podzolica*

* The two species did not visibly interact with each other.

Aphidoidea

The most frequently attended aphids were those of the genera *Aphis*, *Cinara* and *Chaitophorus* (Table 1). *Chaitophorus populicola* Thomas had the greatest number of different ant attendants, followed by *Aphis neogillettei* Palmer, *Myzus cerasi* (Fabricius) and *Chaitophorus nigrae* Oestlund, respectively. Paralleling the extensive distribution of *Chaitophorus populicola* was one of its attendants, *Camponotus noveboracensis* (Tables 1 and 3).

In addition to the finding of Bradley and Hinks (1968) that *Prociphilus* sp. was tended by *A. latipes* in Manitoba, *Prociphilus* was found in the nest of *Lasius pallitarsis* (Provancher) and, in moss, in the presence of *Myrmica emeryana* Forel. *Paraprociphilus tesselatus*, *Thecabius affinis* (Kalt.) and *Pemphigus* sp. were also each collected in association with one ant species.

Host Plants

Populus spp. followed by *Salix* spp. and *Cornus stolonifera* Michx. were the hosts of the greatest numbers of different ant-aphid associations (Table 4). These host plants are widely distributed in Manitoba, and likely occur in the territories of many species of ants. In any given area they probably are the hosts of more different aphid species and more aphid colonies, supplying a larger portion of the area's honeydew than any other plant species.

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Table 4. Number of different ant-aphid associations collected on different host plants in Manitoba, 1976 and 1977.

Host plants	Number of associations
Acer negundo L.	1
Alnus rugosa (Du Roi) Spreng.	1
Amelanchier alnifolia Nutt.	4
Apocynum cannabinum L.	2
Artemisia sp.	3
Artemisia frigida Willd.	2
Betula glandulosa var. *glandulifera* (Regel) Gl.	5 — 1*
Betula papyrifera Marsh.	1 — 1*
Cornus stolonifera Michx.	19
Crataegus sp.	4
Diervilla lonicera Mill.	6
Epilobium angustifolium L.	10
Fraxinus sp.	1
Galium boreale L.	4
Juniperus horizontalis Moench	2
Larix laricina (Du Roi) K. Koch	8
Moss	1
Oenothera biennis L.	5
Philadelphus coronarius L.	2
Picea glauca (Moench) Voss	7
Pinus banksiana Lamb.	14
Populus sp.	10 — 1 —)
Populus balsamifera L.	17 — 8 —) 2
Populus tremuloides Michx.	42 —)
Prunus sp.	2
Prunus pensylvanica L. f.	14
Prunus pumila L.	2 — 1
Prunus virginiana L.	7 — 1
Pteridium aquilinum (L.) Kuhn	1
Quercus macrocarpa Michx.	12
Rosa acicularis Lindl.	2
Rubus strigosus Michx.	3
Rudbeckia hirta L.	1
Salix sp.	25 — 3
Salix bebbiana Sarg.	5 — 3
Salix planifolia Pursh	2
Spiraea alba Du Roi	6
Symphoricarpos occidentalis Hook.	7
Taraxacum officinale Weber	1
Viburnum opulus nanum	1
Viburnum rafinesquianum Schultes	2
Viburnum trilobum Marsh.	1
Arctium minus (Hill) Bernh.	2

*Number of association(s) common to two or more host plants, e.g., the association that occurred on *B. papyrifera* also occurred on *B. glandulosa*.

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