FIELD INCIDENCE OF MOSQUITO PATHOGENS AND PARASITES IN CENTRAL ALBERTA

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ABSTRACT. Ten pools and ponds were monitored on a weekly basis for presence of mosquito pathogens and parasites over a three year period near Edmonton, Alberta. Acari, fungi, Microsporidia and Peritrichida were found associated with mosquitoes. Percentage of collections with pathogens and parasites (prevalence) followed by the mean percentage infection within the samples, in parentheses (estimated % incidence) for the three year period were as follows: *Coelomomyces psorophorae* var. *psorophorae* 0.9(0.01–0.02), *Culicinomyces clavisporus* 1.2(0.09–0.2), Saprolegniales 50.1(15), *Smittium* sp. 23.0(4), Microsporidia 10.9(0.6–1.4), Peritrichida 43.4(30) and Acari 3.2(0.04). The high incidence of Saprolegniales may be a result of attack on stressed individuals under laboratory conditions. Several host and country records are reported. It is concluded that pathogens and parasites generally had little effect on the mosquito populations.

INTRODUCTION

Recently there has been much interest in use of pathogens and parasites as control agents of mosquitoes. Many potential control agents have been identified (see Jenkins 1964, Roberts and Castillo 1980, Roberts and Strand 1977, Roberts et al. 1983), however, most of these represent limited collections and very little is known about their epizootiology. Very few studies have been made to determine fluctuations in parasite activity or the long term incidence of parasites. Such studies are required in order to better evaluate pathogens and parasites as potential biological control agents of mosquitoes. The present study was made to determine importance and seasonal incidence of naturally occurring mosquito pathogens and parasites in 10 selected pools in central Alberta.

MATERIALS AND METHODS

Larval monitoring and diagnosis. Ten pools and ponds 4 km NW of Devon, Alberta (114°47′W, 53°23′N) were regularly monitored between June 1982 and September 1984. In 1982, monitoring began June 15; in 1983 on April 28; and in 1984 on May 6. Five to 120 (usually 10–30) samples were taken from each pool on a weekly basis using a 350 ml dipper. During each field trip, pool parameters such as water temperature (taken 10 cm below the water surface), pH and conductivity were monitored. Dippers and the collectors' boots were rinsed in 5% household bleach after collection at each site in order to minimize the possibility of spreading pathogens between study sites.

Field-collected immatures (i.e., larvae and pupae) were brought back to the laboratory, visually examined for presence of pathogens or parasites, counted and placed into Bates' medium S (McLintock 1952) in trays or 500 ml plastic containers. Immatures were reared at 20°C until emergence or death. Dead larvae were removed daily, identified to species using the key of Wood et al. (1979), and then examined microscopically for signs of pathogens and parasites. At times, dead immatures were stored at 4°C for 24–48 hr prior to examination.

Pathogen/parasite identifications/isolations. Pupae parasitized by mites were removed and placed in a separate container until adult emergence. Adults were then examined for mite infestation. Mites preserved in 70% alcohol were sent to Dr I. M. Smith, Biosystematics Research Centre, Ottawa for identification.

Microsporidian-infected immatures were kept at 4°C in sterile water and sent to Dr A. H. Undeen, Insects Affecting Man and Animals Research Laboratory, Gainesville, FL, for identification. Whole cadavers with *Coelomomyces* infections were mounted on slides in lactofuchsin and identified using the key of Couch and Bland (1985). Immatures infected with fungi other than *Coelomomyces* were bathed for 5 min in 50 μg/ml chloromycetin before being placed on the surface of Sabouraud dextrose agar supplemented with 60 μg/ml penicillin and 30 μg/ml streptomycin. Hyphal growth on the agar was subcultured until pure cultures were obtained. Saprolegniaceous fungi were subcultured onto 2.5% V8® juice with 2% agar and sent for identification to Dr D. J. S. Barr, Biosystematics Research Centre, Ottawa.

Initial attempts were made to isolate pure cultures of most fungal pathogens. However, due to the high incidence of fungi in the order Saprolegniales and difficulties associated in isolation and identification, this was discontinued after the first few months. Subsequently only

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Culicinomyces clavisporus Couch, Romney and Rao was isolated; details are presented elsewhere (Goettel et al. 1984). No attempts were made to isolate trichomycete fungi. These were preserved in lactofuchsin on slides and were sent for identification to Dr R. W. Lichtwardt, Department of Botany, The University of Kansas, Lawrence.

Peritrichida were not identified. No attempts were made to identify or isolate possible bacterial pathogens.

Percent incidence estimates. Since large numbers of mosquitoes were unaccounted for while reared under laboratory conditions and only dead immatures were examined for pathogens and parasites, three methods were used to estimate infection rates of field-collected mosquitoes as follows:

Minimum estimated % incidence
= number infected/total collected × 100. (1)

In this calculation, it is assumed that none of the missing immatures were infected and therefore it estimates the absolute minimum infection rate possible. This calculation was used for *Coelomomyces, Culicinomyces* and Microsporidia. For Acari only the number of mosquito pupae were used in this calculation.

Maximum estimated % incidence
= estimated number infected/total collected × 100. (2)

The estimated number infected was determined by multiplying the proportion of accounted dead immatures with infection by the total number collected, less the number of adults emerged. In this calculation, it is assumed that the same proportion of missing immatures were infected as those that were accounted for. This calculation also takes into account the number of adults emerging. This method was used for *Coelomomyces, Culicinomyces* and Microsporidia.

Estimated % incidence
= number infected/number dead × 100. (3)

In this calculation, it is assumed that the same proportion of all immatures collected were infected as those that died and were accounted for. This calculation was used for Saprolegniales, *Smittium* and Peritrichida.

RESULTS AND DISCUSSION

Habitats. Details of the 10 study sites are summarized in Table 1. Sites A, B, G and I were not monitored in 1982. Sites C and D were dry in 1984 as a result of drainage operations nearby. All other sites were monitored for the entire study period as long as they contained water. Properties of the water were generally similar at all sites. Temperatures of between 3 and 5°C were recorded in early spring. The highest temperature recorded was 25°C at sites H and I. At the other sites, the highest temperatures ranged between 20 and 24°C. Conductivity ranged between 105 and 1710 μmhos/cm and pH between 6.0 and 9.2. In 1982 and 1983, the total rainfall between June and August was approximately 30 cm while in 1984 during the same period it was 5.5 cm (Environment Canada, Edmonton International Airport 14 km SE of the study area). As a result, in 1984 most pools were dry by late summer.

Mosquitoes. In general, pools were first colonized by spring *Aedes* species. Specimens collected included: *Aedes cataphylla* Dyar, *Ae. euedes* Howard, Dyar and Knab, *Ae. excrucians* (Walker), *Ae. fitchii* (Felt and Young), *Ae. flavescens* (Müller), *Ae. mercurator* Dyar, *Ae. pionips* Dyar, *Ae. punctor* (Kirby), and *Ae. riparius* Dyar and Knab. In early June, pools were generally colonized by *Culiseta alaskaensis* (Ludlow) followed later in the summer by *Cs. inornata* (Williston), *Cs. minnesotae* Barr, *Cs. morsitans* (Theobald), and *Culex territans* Walker. *Aedes vexans* (Meigen) colonized sites A, I and J in early to mid-summer.

In the present study identifications were attempted with specimens that had died and were generally in the process of deterioration. Because 4th-instar specimens in excellent condition are required for proper identification of spring *Aedes* species, the identifications are tenuous. Therefore, no attempts are made at establishing new host records for these species.

Many field-collected immatures disappeared while held in the laboratory. This occurred mainly when earlier instars were collected. Of 37,462 immatures collected in 339 collections, 45% were unaccounted for from the time of collection to the time the last individual either died or emerged as an adult. This was probably due to rapid decomposition and cannibalism of immatures held in the laboratory. Predators were also accidentally introduced with the sample in some cases. Of the 55% of immatures that were accounted for, 67% (13,742) died and were diagnosed microscopically for pathogens and parasites.

Pathogens and Parasites. Acari, fungi, Microsporidia and Peritrichida were found associated with mosquitoes (Table 2). The most prevalent (i.e., occurring in most collections) were saprolegniaceous fungi followed by commensal Peritrichida and Trichomycetes, and then microsporidian pathogens. Ectocommensal Peritri-

Table 1. Descriptions of pools in central Alberta monitored for mosquito pathogens and parasites between June 1982 and September 1984.

Site	Habitat description	Vegetation[1]	Size (m)
A	Shallow, semipermanent pool in partially wooded area	*Carex* sp. (d) *Populus* spp. *Salix* sp. (s) *Typha latifolia* (s)	15 × 34
B	Shallow, temporary roadside ditch	*Caltha* sp. (s) *Salix* sp. (s) *Typha latifolia* (s)	1 × 8
C	Deep, semipermanent roadside pool with seepage from adjacent lake	*Lemna minor* *Salix sp.* (s) *Typha latifolia* (s)	25 × 50
D	Same as C	Same as C	30 × 40
E	Shallow, temporary roadside ditch	*Carex* sp. (d) *Typha latifolia* (s)	2 × 25
F	Large shallow marsh fed by stream	*Caltha* sp. *Carex* sp. (d) *Populus balsamifera* (s) *Salix* sp. *Typha latifolia* (s)	210 × 210
G	Large shallow marsh	*Carex* sp. (d) *Salix* sp. (s) *Typha latifolia* (s)	600 × 750
H	Permanent pond	*Carex* sp. (d) *Lemna minor* (d) *Salix* sp. (s)	25 × 50
I	Shallow, temporary roadside pool	*Carex* sp. (d) *Salix* sp. (s) *Typha latifolia* (s)	18 × 25
J	Same as I	Same as I	30 × 50

[1] d = dominant, s = scattered

chida had the highest incidence (Table 2). The least abundant organisms were Acari, *Coelomomyces* and *Culicinomyces*.

Incidences of pathogens and parasites were generally too low to make any comparisons between pools and years. There were no apparent correlations between pool parameters and infection rates. Details of pool parameters are therefore presented only for *C. clavisporus* as very little is known about its occurrence in nature.

FUNGI

Coelomomyces. Only 4 larvae were found infected with *Coelomomyces* on 3 occasions and at 2 sites. All were identified as *Coelomomyces psorophorae* var. *psorophorae* Couch with *Ae. vexans* as the only host. The first infection was detected at site J on July 23, 1982. This pool was flooded as a result of heavy rain in the first week of July and was colonized by large numbers of *Ae. vexans*. The sample consisted of 11 dips which yielded 581 larvae (primarily fourth-instar) and 43 pupae. The infected larva died 3 days post-collection.

The second and third occurrences were detected on June 28 and July 12, 1983 at site A. On June 28, 44 larvae (3 first, 35 second, 6 third-instar) were collected in 10 dips and on July 12, 33 larvae (8 first, 11 second, 14 third) and 50 pupae were collected in 5 dips. The single infected larva from the June 28 collection died 17 days post-collection while the 2 larvae from the July 12 collection died 6 and 9 days post-collection.

Coelomomyces psorophorae var. *psorophorae* has been collected from many mosquito hosts and habitats world-wide (Couch and Bland 1985). In Canada *C. psorophorae* var. *psorophorae* is widely distributed and well established in southern Alberta occurring in larvae of *Cs. inornata* and less commonly in *Ae. vexans* (Shemanchuk 1959, Zebold et al. 1979). Incidences of up to 80% have been observed in *Cs. inornata* (Shemanchuk 1977) while in 1956, 12% of all *Cs. inornata* were infected (Shemanchuk 1959). *Coelomomyces* has also been reported from *Ae. trivittatus* (Coq.) in Manitoba (Taylor et al. 1980). These authors reported incidence rates of up to 56% in field-collected mosquitoes, however, infections were apparent in blood-fed adults only. Infection rates in the present study may therefore have been higher since adults were not examined. In the Manitoba study how-

Table 2. Summary of mosquito pathogen and parasite prevalence and incidence in 10 study sites in central Alberta.

Site/year	No. collected	Prevalence[1] (estimated % incidence)[2]											
		FUNGI								MICROSPORA Microsporidia		CILIOPHORA Peritrichida		ACARI	
		Coelomomyces		*Culicinomyces*		Saprolegniales		*Smittium*							
A															
1983	874	20	(2–5)	—		50	(39)	30	(12)	20	(2–4)	60	(74)	—	
1984	348	—		9	(1)	54	(73)	—		—		54	(86)	—	
B															
1983	604	—		—		57	(37)	43	(6)	—		71	(79)	—	
1984	1002	—		—		83	(66)	8	(1)	—		83	(79)	—	
C															
1982	819	—		—		50	(71)	12	(1)	19	(4)	6	(1)	19	(6)
1983	211	—		—		40	(80)	—		—		13	(64)	—	
D															
1982	540	—		—		33	(19)	—		5	(1–5)	—		17	(5)
1983	121	—		—		8	(60)	—		—		17	(69)	—	
E															
1982	3569	—		—		56	(45)	56	(12)	36	(12–31)	9	(6)	9	(2)
1983	1301	—		—		57	(45)	29	(24)	5	(1–2)	43	(69)	—	
1984	129	—		—		75	(42)	—		—		100	(100)	—	
F															
1982	878	—		—		19	(9)	6	(12)	12	(10)	6	(8)	12	(9)
1983	2460	—		—		76	(53)	48	(27)	9	(4–5)	81	(77)	—	
1984	573	—		—		64	(83)	14	(2)	—		71	(79)	—	
G															
1983	632	—		5	(26–38)	61	(33)	22	(8)	—		67	(95)	—	
1984	584	—		—		53	(95)	—		—		60	(82)	—	
H															
1982	961	—		10	(6–16)	26	(26)	16	(25)	—		—		5	(2)
1983	349	—		—		40	(48)	—		13	(2–3)	40	(66)	—	
1984	334	—		—		57	(61)	14	(4)	—		71	(68)	—	
I															
1983	2992	—		—		33	(41)	33	(13)	7	(10)	40	(70)	—	
1984	2165	—		—		61	(90)	23	(3)	8	(3)	78	(88)	—	
J															
1982	4438	10	(0.2–0.5)	—		60	(43)	70	(49)	80	(3–10)	—		—	
1983	2832	—		—		61	(79)	61	(16)	23	(19–30)	77	(75)	—	
1984	8746	—		—		67	(20)	60	(8)	—		93	(83)	—	
Total[3]	37462	0.9	(0.01–0.02)	1.2	(0.09–0.2)	50.1	(15)	23.0	(4)	10.9	(0.6–1.4)	43.4	(30)	3.2	(0.3)

[1] Percent of collections where mosquitoes were found.

[2] Maximum % incidence in field collected immatures reared under laboratory conditions; for *Coelomomyces*, *Culicinomyces*, and Microsporidia = no. infected/total collected × 100 to no. estimated infected/total collected × 100; for Saprolegniales, *Smittium* and Peritrichida = no. infected/no. dead × 100; for Acari % incidence = no. infected/total no. of pupae collected at the site for the year × 100; see text for details.

[3] Pooled data.

ever, there was no evidence of infection in any blood-fed *Ae. vexans* that were collected from the same pools where infected *Ae. trivittatus* were found. In related studies, adults of *Ae. sticticus* (Meigen) were found infected (loc. cit.). There are only 2 other records of *Coelomomyces* occurring in Canada; *C. borealis* var. *giganteus* Couch and Bellamy collected in Ontario in *Ae. fitchii* (Felt and Young) and *Ae. stimulans* (Walker) and *C. canadensis* (Weiser and McCauley) Nolan collected from a chironomid larva in British Columbia (Couch and Bland 1985).

Culicinomyces. Culicinomyces clavisporus was detected in every study year. The first infections occurred in collections of August 12 and 19, 1982 at site H (Goettel et al. 1984).

The second occurrence of *C. clavisporus* was found in the collection of July 26, 1983 from site G. Properties of the water were 17°C, 8.3 pH, and 550 μmhos/cm conductivity. Thirty dips yielded 69 larvae (50 first, 14 second, 4 third, and 1 fourth-instar). Of 45 larvae identified, 51% were *Cs. inornata* and the remainder were *Cx. territans*. A total of 17 infected larvae of *Cs. inornata* died between 7 and 22 days post-collection. A single infected larva of *Cx. territans* was found 23 days post-collection. A pure culture was obtained from this site and has been deposited at the University of Alberta Microfungus Collection and Herbarium as UAMH 4854.

The third occurrence of *C. clavisporus* was found in the collection of June 12, 1984 from site A. Properties of the water were 12°C, 7.3 pH and 600 μmhos/cm conductivity. Thirty dips yielded 259 second instar larvae of *Ae. vexans*. Two infected larvae died 5 days post-collection. On the following weekly collections, only 1 larva and 4 pupae were collected from this site before it dried on July 3 for the rest of the study period.

The recovery of *C. clavisporus* in Canada from a permanent pond, a marsh and a semi-permanent pool broadens the range of its known aquatic habitats (rockpools, streams, ponds and lakes) and its geographic distribution (USA, Australia, Canada). *Culiseta inornata* and *Ae. vexans* are new records for mosquito hosts infected in nature; since the single infected *Cx. territans* larva died 23 days post-collection, this was probably a laboratory acquired infection. This species was previously reported susceptible in laboratory challenge tests (Couch et al. 1974).

Saprolegniales. Fungi in the order Saprolegniales were the most prevalent, occurring in 50% of all collections. Incidences of up to 95% of all dead larvae were recorded and infections were noted in virtually all species collected. Initial identifications included *Saprolegnia ferax* (Gruith) Thuret from spring *Aedes, Cs. alaskaensis* and *Ae. vexans* and *S. hypogyna* (Pringshein) deBary from larvae of *Cs. alaskaensis* and from an unidentified mosquito pupa. Other occurrences noted as Saprolegniales may have included species in the genera *Achlya* and *Aphanomyces*, however isolations of these species were not made.

Saprolegnia species have been isolated from mosquito larvae on many occasions (Kalvish and Kukharchuk 1974, Roberts and Strand 1977), however, it is generally believed that these are infections of weakened or dying larvae. Even though laboratory studies have demonstrated high mortalities (i.e., Rioux and Achard 1956), little is known of the potential of *Saprolegnia* species as biocontrol agents of mosquitoes.

This is the first record of *S. ferax* in mosquitoes. *Culiseta alaskaensis* is a new host record for *S. hypogyna*. Previously it was reported from *Ae. excrucians* in the USSR (Kalvish and Kukharchuk 1974).

Trichomycetes. Many larvae were infected with the ectozootic *Amoebidium parasiticum* Cienk, however, since there is no reported evidence that it is detrimental to mosquitoes, details of its prevalence were not recorded. Many dead larvae were also infected with a *Smittium* species. Examination of slide-mounted larvae of *Ae. vexans* and *Cs. inornata* revealed that the fungus was probably *Smittium culisetae* Lichtwardt, however, Dr Lichtwardt noted "there is some question about the type of branching and the way the spores are borne on the fertile tips". I therefore refer to this fungus as *Smittium* sp. Larvae were found infected in up to 70% of the collections (i.e., at site J in 1982) with up to 49% of dead larvae infected.

Trichomycetes are widely distributed and live obligately within the digestive tracts of arthropods with the exception of the Amoebidiales which live externally (Lichtwardt 1976). Trichomycetes of the genus *Smittium* were not generally thought to be detrimental to their hosts, however, high mortalities resulted after first-instar larvae of *Ae. aegypti* (Linn.) were fed large numbers of *S. culisetae* spores (Williams and Lichtwardt 1972). It was speculated that this was a result of poor nutrition of the host. Recently Sweeney (1981) isolated *S. morbosum* Sweeney which was responsible for mortality rates of 50–90% in laboratory colonies of *An. hilli* Woodhill and Lee. *Smittium morbosum*, unlike other species in this genus, was found to penetrate the midgut cells and at times the cells of Malpighian tubules. Others also report mortalities in mosquitoes as a result of *Smittium* infections (in Sweeney 1981).

In the present study, diagnoses of *Smittium* sp. were made only from dead larvae. It is there-

fore not known what proportion of the population was infected or if the fungus was a factor contributing to mortality. Diagnoses were most often made by observing a sporulating thallus protruding from the anus of the cadaver. This was also observed by Sweeney (1981) in cadavers infected with *S. culisetae*. Since I made no dissections, the estimated incidence rate is probably conservative. There was no evidence of infections by *S. morbosum* (i.e., blackened appearance of invasion sites along the midgut).

In the present study *Smittium* sp. occurred in virtually all mosquito species collected with most observations in *Ae. vexans* followed by *Cs. inornata* (Table 3). This is the first report of a *Smittium* sp. from mosquitoes in Canada; *Cs. alaskaensis, Cs. minnesotae, Cs. morsitans* and *Cx. territans* are new host records; it has also not been previously reported from any of the spring *Aedes* species that occur in the Edmonton area. It has been previously recorded from *Ae. vexans* and *Cs. inornata* in Nebraska (Williams and Nagel 1980).

The only previous study conducted on the field incidence of *Smittium* species in mosquitoes was a 2 year survey in Nebraska by Williams and Nagel (1980). They found *Smittium* occurring most frequently in *Cs. inornata* with an annual infection rate of up to 53%; only low infection rates occurred in *Ae. vexans* (4%). The present results further support their observations that *Smittium* does not appear to be host specific. Williams and Nagel also speculated that *Smittium* was dependent upon the continued presence of hosts to maintain a population. The results of this study indicate otherwise. *Smittium* was commonly found in *Ae. vexans* and occurred in temporary pools shortly after they were flooded and had previously been dry for extended periods.

Table 3. Summary of *Smittium* sp. and Microsporidia occurring in 10 study sites in central Alberta between June 1982 and September 1984.

Host	Number of specimens collected	
	Smittium sp.	Microsporidia
Spring *Aedes*	10	27
Ae. vexans	287	113
Cs. alaskaensis	46	3
Cs. inornata	93	16
Cs. minnesotae	4	8
Cs. morsitans	2	5
Cx. territans	9	16
Unidentified	41	43
Total	491	231

MICROSPORA

Microsporidia. Microsporidians occurred at 8 of the 10 study sites. Prevalence was highest in 1982, however in 1984 only a single specimen was found infected (collected on May 1 at site I). Highest estimated incidence was 19–30%, however overall incidence over the three years was under 2%.

There were difficulties in identifications of microsporidians since diagnoses were made only after larvae had died. Most microsporidians were identified as *Amblyospora* species possibly *A. inimica* (Kellen and Wills) Hazard and *A. opacita* (Kudo) Hazard. Since it was not possible to obtain identifications for the majority of the infected specimens, I refer to these as Microsporidia.

Microsporidian parasites occur worldwide, infecting well over 100 mosquito species (Roberts and Castillo 1980, Roberts et al. 1983, Roberts and Strand 1977). Field infections of Microsporidia are usually 1% or less, however, epizootics of 80 to 99% incidence are known (Andreadis 1983, Chapman 1974).

Species found infected in the present study are presented in Table 3. New host records for Microsporidia include *Cs. alaskaensis, Cs. minnesotae* and *Cs. morsitans*. Previous records include *Ae. vexans, Cs. inornata, Cx. territans* and many *Aedes* species that occur in the Edmonton area in the spring; *Parathelohania* sp. occurring in *An. earlei* Vargas and *Nosema* sp. occurring in *Ae. excrucians* in Quebec and *A. khaliulini* Hazard occurring in *Ae. communis* (DeGeer) at Churchill, Manitoba are the only previous Canadian records (Roberts and Castillo 1980, Roberts et al. 1983, Roberts and Strand 1977).

CILIOPHORA

Peritrichida. Peritrichs were abundant in most collections with up to 100% of dead larvae being infested and they occurred on virtually all species collected. The species involved was probably *Vorticella* sp.

Peritrichida occur frequently as epibionts on mosquito larvae and are usually considered not detrimental to their "host". There are, however, numerous reports of detrimental effects, including apparent mortality rates of up to 100% (Roberts and Strand 1977, Jenkins 1964). Canadian records of peritrichs on mosquitoes include Manitoba, Ontario and Quebec (see Welch 1960 and references therein).

ACARI

Acari. Mites were found only in 1982 occurring in 5 out of the 6 sites monitored that year.

The first occurrence of mites was on June 24 at site E at which time a single spring *Aedes* sp. pupa was collected with 156 mites attached. These immatures were identified as belonging to the genus *Arrenurus* by Dr I. M. Smith. Mites encountered subsequently were not identified. The prevalence for 1982 was 11% with an estimated incidence rate of 0.5% (expressed as number of pupae infected/total number of pupae collected).

Mosquito-parasitizing mites have a wide geographical distribution and *Arrenurus* spp. are the most common (Mullen 1975). It is generally believed that mites reduce fecundity and longevity of their host and therefore have biological control potential (Smith 1983). Larvae of *Arrenurus* spp. are common in the tropics and subtropics and in some populations the prevalence of parasitization can reach 80%. In temperate regions mosquitoes are generally parasitized only occasionally, but prevalence within a population may be high (see references in Smith 1983).

In the present study, mites were found infecting spring *Aedes* species, *Cs. inornata, Cs. minnesotae* and *Cx. territans; Cs. minnesotae* is a new host record. Previously mites were found on spring *Aedes* species and *Cs. inornata* in Alberta and on *Cx. territans* in Quebec (Leprince 1981, Mullen 1975).

Pathogens not found. It is interesting that neither viruses nor nematodes were found. There are no records of viruses from mosquito larvae in Canada, however they are probably distributed worldwide (Federici 1985). Incidences of virus disease in nature are usually less than 1%, but epizootics with infection rates of up to 70% occur (Federici 1985). It is highly unlikely that in the present study infections of iridoviruses were overlooked. This disease is one of the easiest to diagnose as infected larvae are iridescent. Several suspect specimens were sent to Dr B. A. Federici, University of California, Riverside and were confirmed as being virus-free.

There are numerous records of nematodes from mosquitoes in Canada (Roberts et al. 1983, Roberts and Strand 1977, Roberts and Castillo 1980) and most infections are also easily diagnosed. It can be concluded that viruses and pathogens were either totally absent from the 10 sites during the study period or occurred at such a low level that they were not detected.

Effects on host population. It is difficult to assess the field incidence of pathogens and parasites and their impact on the mosquito population especially when they occur at very low levels. Specific difficulties encountered in the present study include the following: (1) Transfer of field-collected immatures to laboratory conditions probably stresses the individuals as evidenced by the high mortality rates witnessed. Stressed individuals may become targets for such "pathogens" as *Saprolegnia.* Most incidences of Saprolegniales were probably the result of such attacks; experiments with controls are required before the potential of these fungi as control agents is ruled out. (2) Since immatures were held in the laboratory, infections may have been acquired in the laboratory. As discussed above, this probably occurred with Saprolegniales since many cadavers were observed while the fungus was releasing zoospores. Some *C. clavisporus* infections were also probably laboratory-acquired as larvae died of infection up to 23 days post-collection. (3) Large numbers of immatures disappeared between the time of collection and the time the last individual either died or emerged as an adult.

In order to estimate incidence in the field as accurately as possible taking the above problems into consideration, 3 methods were used. Methods 1 and 2 were used for *Coelomomyces, Culicinomyces* and Microsporidia to give an estimated range of percent incidence. It is assumed that the possibility of laboratory acquired infections is minimal (in the case of *Coelomomyces* and *Culicinomyces*, the numbers found were so low that laboratory acquired infections would have little effect on the estimates in any case). Method 3 was used for *Smittium* sp. and Peritrichida as only dead larvae were diagnosed. Since these organisms are generally considered not detrimental to their "host", it can be assumed that the same proportion of all immatures collected were infected as those that died and were accounted for. If on the other hand, these organisms contributed to the death of the mosquitoes, this is an inaccurate method for estimating field incidence.

Concluding remarks. Even though difficulties were encountered in accurately estimating incidence in the field, pathogens and parasites appear to have had little effect on the larval mosquito populations studied. Similar observations were made by Service (1977) in England. In a 6-year study of *Ae. cantans,* he estimated that although 95% larval and pupal mortality occurred, few were killed by pathogens and parasites.

The many new locality and host records reported in the present study further demonstrate that the known geographical distributions of pathogens and parasites are only a reflection of the geographic distribution of entomologists interested in pathogens and parasites of mosquitoes (Chapman 1974). The extremely low prevalence and incidence of some pathogens dem-

onstrates how difficult it is to establish such records unless long term studies are undertaken. In terms of control of mosquitoes, the wide geographical distribution and low incidence of pathogens and parasites in nature indicates that the inoculative method of biological control may not be successful. Therefore inundative use of these pathogens and parasites will probably be required. Further, if parasites and pathogens are ever to be fully exploited for mosquito control, a much better understanding of biotic and abiotic conditions causing epizootics is necessary.

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REFERENCES CITED

Andreadis, T. G. 1983. An epizootic *Amblyospora* sp. (Microspora: Amblyosporidae) in field populations of the mosquito *Aedes cantator*. J. Invertebr. Pathol. 42:427–430.

Chapman, H. C. 1974. Biological control of mosquito larvae. Annu. Rev. Entomol. 19:33–59.

Couch, J. N. and C. E. Bland. 1985. Taxonomy. pp. 81–297. *In:* J. N. Couch and C. E. Bland (eds). The Genus *Coelomomyces*. Academic Press, New York.

Couch, J. N., S. V. Romney and B. Rao. 1974. A new fungus which attacks mosquitoes and related Diptera. Mycologia 66:374–379.

Federici, B. A. 1985. Viral pathogens. pp. 62–74. *In:* H. C. Chapman (ed). Biological control of mosquitoes. Am. Mosq. Control Assoc. Bull. 6.

Goettel, M. S., L. Sigler and J. W. Carmichael. 1984. Studies on the mosquito pathogenic hyphomycete *Culicinomyces clavisporus*. Mycologia 76:614–625.

Jenkins, D. 1964. Pathogens, parasites and predators of medically important arthropods: Annotated list and bibliography. Bull. W. H. O. 30 (Suppl).

Kalvish, T. K. and L. P. Kukharchuk. 1974. Pathogenic mycoflora of blood-sucking mosquitoes of Western Siberia and the Far East. Med. Parazitol. Bolezni 43:57–64. (in Russian with English summary).

Leprince, D. J. 1981. Inventaire prèliminaire d'acariens (Acari:Hydrophantidae et Arrenuridae) ectoparasites de moustiques (Diptera: Culicidae) du lac Boivin, Granby, Quebec. Can. J. Zool. 59:8–18.

Lichtwardt, R. W. 1976. Trichomycetes. pp. 651–671. *In:* E. B. Gareth Jones (ed). Recent advances in aquatic mycology. Elek Science, London.

McLintock, J. 1952. Continuous laboratory rearing of *Culiseta inornata* (Wilt.) (Diptera: Culicidae). Mosq. News 12:195–201.

Mullen, G. A. 1975. Acarine parasites of mosquitoes. I. A critical review of all known records of mosquitoes parasitized by mites. J. Med. Entomol. 12: 27–36.

Rioux, J. A. and F. Achard. 1956. Entomophytose mortelle a *Saprolegnia declina* Humphrey 1892 dans un elevage d' *Aedes berlandi* Seguy 1921. Vie et Milieu 7:326–337.

Roberts, D. W. and J. M. Castillo (eds). 1980. Bibliography on pathogens of medically important arthropods: 1980. Bull. W. H. O. 58 (Suppl. 1).

Roberts, D. W., R. A. Daoust and S. P. Wraight. 1983. Bibliography on pathogens of medically important arthropods: 1981. W. H. O. VBC/83.1.

Roberts, D. W. and M. A. Strand (eds). 1977. Pathogens of medically important anthropods. Bull. W. H. O. 55 (Suppl. 1).

Service, M. W. 1977. Ecological and biological studies on *Aedes cantans* (Meig.) (Diptera: Culicidae) in southern England. J. Appl. Ecol. 14:159–196.

Shemanchuk, J. A. 1959. Note on *Coelomomyces psorophorae* Couch, a fungus parasitic on mosquito larvae. Can. Entomol. 91:743–744.

Shemanchuk, J. A. 1977. Biological control of mosquitoes. Canada Agric. 22(4):25–26.

Smith, B. P. 1983. The potential of mites as biological control agents of mosquitoes. pp. 79–85. *In:* M. Hoy, G. Cunningham and L. Knutson (eds). Research needs for development of biological control of pests by mites. Agric. Exp. Stn. Univ. Calif. Spec. Publ. No. 3304.

Sweeney, A. W. 1981. An undescribed species of *Smittium* (Trichomycetes) pathogenic to mosquito larvae in Australia. Trans. Br. Mycol. Soc. 77:55–60.

Taylor, B. W., J. A. Harlos and R. A. Brust. 1980. *Coelomomyces* infection of the adult female mosquito *Aedes trivittatus* (Coquillett) in Manitoba. Can. J. Zool. 58:1215–1219.

Welch, H. E. 1960. Effects of protozoan parasites and commensals on larvae of the mosquito *Aedes communis* (DeGeer) (Diptera: Culicidae) at Churchill, Manitoba. J. Invertebr. Pathol. 2:389–395.

Williams, M. C. and R. W. Lichtwardt. 1972. Infection of *Aedes aegypti* larvae by axenic cultures of the fungal genus *Smittium* (Trichomycetes). Am. J. Bot. 59:189–193.

Williams, M. C. and H. G. Nagel. 1980. Occurrence of Trichomycete fungi in mosquito larvae near Kearney, Nebraska. Mosq. News 40:445–447.

Wood, D. M., P. T. Dang and R. A. Ellis. 1979. The mosquitoes of Canada. 390 p. Part 6. *In:* The insects and arachnids of Canada. Biosystematics Research Institute, Ottawa. Publ. 1686.

Zebold, S. L., H. C. Whisler, J. A. Shemanchuk and L. B. Travland. 1979. Host specificity and penetration in the mosquito pathogen *Coelomomyces psorophorae*. Can. J. Bot. 57:2766–2770.