

Myiasis by *Lucilia silvarum* (Calliphoridae) in Amphibian Species in Boreal Alberta, Canada

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ABSTRACT: We report myiasis by *Lucilia silvarum* with an overall prevalence of 0.9% in amphibian populations in boreal Alberta. In the period 1998–1999, we documented *L. silvarum* infestations in wild populations of wood frog (*Rana sylvatica*), boreal chorus frog (*Pseudacris maculata*), boreal toad (*Bufo boreas boreas*), and Canadian toad (*B. hemiophysys*). We believe this is the first record of this parasite from boreal and Canadian toads. Almost all previous records of *L. silvarum* parasitism in North America indicate that myiasis is fatal to an anuran host. Here, we provide the first record of adult individuals from 2 species (wood frog and boreal toad) surviving infestations. Although we actively captured and examined amphibians in Alberta from 1996 to 2006, we only found parasitism in 1998 and 1999. This is the most northerly record of anuran infestations by this parasite.

Two green blowfly species (Calliphoridae: *Lucilia*) are known to cause myiasis in anurans in North America: *Lucilia silvarum* and *L. elongata* (Baumgartner, 1988) (note that formerly these species were sometimes placed in *Bufo lucilia* (Whitworth, 2006)). The limited records available suggest that *L. silvarum* is more common than *L. elongata*, and it is the only member of the genus that has been recorded from Alberta, Canada, the site of this study (Strickland, 1938; Roberts, 1998; for a description of *L. silvarum* see Hall, 1948 and Whitworth, 2006).

Adult green blowflies deposit eggs on the backs and flanks of healthy anurans (Bolek and Janovy, 2004). Larvae hatch within hours of deposition, burrow into the skin, and travel subcutaneously to congregate in a single lesion. The larvae grow rapidly and in less than 1 wk, third-stage maggots leave the amphibian's body, burrow into the soil and pupate. Adult blowflies usually emerge within 9 days of pupation (Baumgartner, 1988; Bolek and Coggins, 2002; Bolek and Janovy, 2004). The life cycle appears to proceed more quickly in ranids than in bufonids (Bolek and Janovy, 2004). *Lucilia silvarum* infestations are usually fatal to the amphibian host (Table I).

Both North American species of *Lucilia*, and their European counterpart, *L. bufonivora*, attack species of *Bufo*. However, in North America, *L. silvarum* has been reported not only in American toad (*Bufo americanus*; Anderson and Bennett, 1963; Bleakney, 1963; Briggs, 1975; Bolek and Coggins, 2002) and boreal toad (*B. boreas boreas*; James and Maslin, 1947), but also in wood frogs (*Rana sylvatica*; Bolek and Janovy, 2004), bullfrogs (*R. catesbeiana*; Hall, 1948), and boreal chorus frogs (*Pseudacris maculata*; Roberts, 1998) (see Table I for a summary of previous records of *Lucilia* spp. attacking amphibians in North America).

Here, we report occurrence and prevalence (defined following Bush et al., 1997) of *L. silvarum* in amphibian species in the boreal mixed wood forest of Alberta, Canada. We performed a systematic survey of amphibian populations in the South Calling Lake area (SCL; 55°07'N, 113°43'W) from 1996 to 1999, and in the Lac La Biche area (LLB; 55°08'N, 111°45'W) in 1996, 1997, and 1999. We sampled amphibians at multiple sites in each area using drift fence and pitfall arrays and visual surveys near lakes, ponds, and in upland habitat (Hannon et al., 2002; Macdonald et al., 2006). We trapped each year between 12 May and 18 July to target adult amphibians (≥ 40 mm snout-to-urostyle length [SUL]; Berven, 1990; Leclair et al., 2000) and yearlings (≤ 30 mm SUL; B. R. Eaton and C. A. Paszkowski, unpubl. obs.), as well as between 20 July and 2 September to target metamorphs (≤ 27 mm SUL; B. R. Eaton and C. A. Paszkowski, unpubl. obs.).

Pitfall traps consisted of a metal can (25.5 cm deep, 15.5 cm diameter) buried to its rim with a plastic funnel (10-cm diameter opening) inside the can. Upright sticks placed in traps provided escape routes for small mammals. The configuration of drift fence–pitfall trap arrays varied with location. Around lakes, each array consisted of 3 pitfall traps joined by two 5-m lengths of 15-cm high plastic drift fence to form a

“V,” with the open side facing the lake. A number of small ponds were entirely surrounded with plastic drift fence 30 cm high, with pitfall traps placed in pairs and with one trap on either side of the fence at 10-m intervals. At upland sites, arrays consisted of 3 drift fence arms (4.5 m long and 30–50 cm high, constructed of plastic or silt fencing) radiating from a central point, with an angle of 120° between arms. Arrays included 6 traps, 3 at the end of the arms and 3 where the arms converged in the center.

We checked the pitfall traps every 2–10 days and identified, measured SUL, weighed, marked, and released any captured amphibians that were unparasitized. We also collected, examined, and released animals encountered during visual searches and incidentally during other activities; unparasitized animals were released at the capture site. Parasitized animals were retained for observation and to rear parasites.

Infested amphibians were held outdoors in plastic containers at ambient temperature in shaded areas. Each container was covered with a piece of window screening so that air could circulate, but flies could not escape. In some cases, especially when the infestation was heavy or with very small animals, e.g., metamorphs, the entire amphibian was consumed before the maggots were large enough to pupate. In these cases, dead amphibians (previously collected and frozen) or fresh beef liver were supplied to the maggots.

Usually, a substratum of sand was provided for the maggots to crawl into and pupate; in some cases, no substratum was supplied, but third-stage maggots were collected from the containers with the dead amphibian and preserved or placed in containers of sand to pupate. Occasionally pupae were found in containers without a substratum; in these cases the larvae appear to have pupated in the carcass of the amphibian. Pupae were placed in a substratum of fine wood chips in small vials, or were left in containers with a sand substratum, until emergence. Adult flies were allowed to die in the containers in which they pupated; they were identified at the Strickland Museum of Entomology at the University of Alberta by D. Shpeley, and 6 specimens (accession numbers UASM97001–UASM97006) were deposited in that museum. Collection and handling procedures for amphibians were in accordance with guidelines from the Canadian Council on Animal Care; protocols were reviewed and approved by the BioSciences Animal Policy and Welfare Committee at the University of Alberta.

In 1998, when we sampled only the South Calling Lake area, we found myiasis in boreal toad (1 adult and 5 juveniles), boreal chorus frog (2 adults), and wood frog (8 adults), with prevalence values ranging from 1.6% (adult wood frog) to 12.8% (juvenile boreal toad; see Table II). The prevalence in all anurans was 2.3%. Mortality of parasitized animals ranged from 0% for adult boreal toad (the only infested animal survived) to 100% for juvenile boreal toad and adult boreal chorus frog (Table II). Mean intensity of these infestations ranged from 3 to 11 parasites per animal (Table II).

All the amphibians included in the data above were found alive; we also found a parasitized dead adult wood frog in a pitfall trap; there were approximately 73 eggs in 3 different locations (right lateral side of head, upper left thigh, lower left flank) on this individual. Because this individual was likely killed by a shrew before it was parasitized, we have excluded it from summary calculations.

In 1999, we sampled both the South Calling Lake and Lac La Biche areas. We documented myiasis in the South Calling Lake area in boreal toad (1 adult and 5 juveniles), and in the Lac La Biche area in Canadian toad (1 adult) and wood frog (1 adult); prevalence ranged from 0.35% (adult boreal toad) to 9.1% (adult Canadian toad; see Table II), with a prevalence for all anurans of 0.4% in South Calling Lake, and 1.2% in Lac La Biche. Mortality was 100% for all boreal and Canadian toads; the only infested wood frog escaped and its fate is unknown (Table II). Mean intensity ranged from 2 to 20 parasites per animal (Table II).

Over the 2 yr we found parasites, we successfully reared 9 adult flies. We were able to identify 6 of these flies; all were *L. silvarum*. Given

TABLE I. Previous records of the green blowfly (*Bufolucilia* sp.) infesting amphibians in North America.

Location	Host species	Life stage	Number examined	Number infected	Mean intensity	% Prevalence	% Death rate	Reference
<i>Bufolucilia elongata</i>								
Wisconsin	<i>Bufo americanus</i>	Adult	59	6	10.8	10.2	NR	Briggs, 1975
Colorado	<i>B. boreas boreas</i>	Adult	1	1	86	NA	NA	James and Maslin, 1947
Wisconsin	<i>Rana sylvatica</i>	Juvenile	39	1	14	2.5	100	Bolek and Janovy, 2004
<i>Bufolucilia silvarum</i>								
Ontario	<i>B. americanus</i>	Adult	1	1	45	NA	100	Anderson and Bennett, 1963
Nova Scotia	<i>B. americanus</i> †	Adult	1	1	15	NA	100	Bleakney, 1963
Wisconsin	<i>B. americanus</i>	Juvenile	140	9	10.5	6.4	100	Bolek and Coggins, 2002
Wisconsin	<i>B. americanus</i>	Adult	10	0	NA	0	NA	Bolek and Coggins, 2002
Alberta	<i>Pseudacris maculata</i>	Adult	3	3	NR	NA	100	Roberts, 1998
Alberta	<i>R. sylvatica</i> ‡	Adult	1	1	NR	NA	100	Roberts, 1998
Wisconsin	<i>R. sylvatica</i>	Juvenile	39	2	29.5	5.1	100	Bolek and Janovy, 2004
California	<i>R. catesbeiana</i>	Adult	59	48	NR	81.3	45.8§	Hall, 1948

* NR = not recorded; NA = not applicable.

† Reported as *B. terrestris americanus*, which is a synonym of *B. americanus*.

‡ No parasites were successfully reared from this individual but material reared from a specimen of *P. maculata* from the same site and time period was identified as *B. silvarum* (Roberts, 1998).

§ Hall (1948) reported on the infestation of *R. catesbeiana* secondhand. Of the 59 bullfrogs examined, 22 were dead; 26 living specimens had maggot infestations in one (15 animals) or both eye sockets (11 animals). No information was provided on whether the infested animals eventually died or not, so the reported death rate is conservative.

the similarity of eggs, larvae, and pupae observed on amphibians, we are assuming that all infestations were attributable to this species.

The mean intensity of infestations in our study was 8.25, which is lower than that previously reported (Table I). *Lucilia silvarum* prevalence in our study was also lower than reported for other populations of North American amphibians, possibly because we collected samples over numerous locations within each study area, and over multiple years, and had sample sizes substantially higher than in most previously published studies (Table I). Given that the active season for both amphibians and dipterans is relatively short at higher latitudes, such as our study area, it is likely that fewer generations of flies are produced an-

nually. Therefore, parasitic fly populations in the north may not reach the abundance that has been documented in locations farther south, where adult flies are active from May to October (Dicke and Eastwood, 1952; Schoof et al., 1956; Judd, 1956, 1958). We captured parasitized amphibians from 27 June to 24 August in 1998, and from 22 July to 22 August in 1999.

The mean host mortality in our study (78.9%; of 19 individuals whose fate was known, 14 died) was lower than most previous studies, where 100% mortality was usual among parasitized individuals (Table I). The only previous case in which mortality was not 100% occurred in bullfrogs, where 45.8% of parasitized animals were found dead (Hall,

TABLE II. Summary of *B. silvarum* infestations from present study.

Year	Location	Host species	Life stage	Number examined	Number infected	Mean intensity†	% Prevalence	% Death rate
1998	South Calling Lake area	<i>Rana sylvatica</i>	Adult	439	8‡	7.0 (8.5; 2)	1.6	25§
			Juvenile	106	0	NA	0	na
		<i>Pseudacris maculata</i>	Adult	37	2	3.0 (1)	5.4	100
			Juvenile	10	0	NA	0	na
		<i>Bufo boreas</i>	Adult	20	1	11.0 (1)	5.0	0
			Juvenile	39	5	3.3 (1.3; 5)	12.8	100
1999	South Calling Lake area	<i>R. sylvatica</i>	Adult	285	0	NA	0	na
			Juvenile	796	0	NA	0	na
		<i>P. triseriata</i>	Adult	21	0	NA	0	na
			Juvenile	1	0	NA	0	na
		<i>B. boreas</i>	Adult	285	1	2.0 (1)	0.35	100
			Juvenile	265	5	13.8 (6.8; 5)	1.9	100
	Lac La Biche area	<i>R. sylvatica</i>	Adult	111	1	20.0 (1)	0.90	Unknown
			Juvenile	30	0	NA	0	na
		<i>P. triseriata</i>	Adult	6	0	NA	0	na
			Juvenile	3	0	NA	0	na
<i>B. hemiophrys</i>	Adult	11	1	5.0 (1)	9.1	100		
	Juvenile	2	0	NA	0	na		

* NA = not applicable.

† Mean intensity = the number of parasites per infested animals divided by the total number of infested animals (standard deviation; sample size).

‡ 1 infested animal found dead was excluded from summary statistics.

§ 3 parasitized wood frogs that were released were not included in calculating death rate.

1948). This information was reported anecdotally and the fate of the remaining parasitized frogs (each of which had maggots infesting at least 1 eye) was not recorded.

The 4 individuals that survived fly infestations, i.e., 3 wood frogs and 1 boreal toad, were all adults. Wounds on the wood frogs healed and the animals were released at their capture sites. The boreal toad had severe infestations over the spine and in the right thigh. Both wounds healed after maggots emerged to pupate, but the animal was retained in captivity. Ours are the first observations of these 2 species surviving infestations by *Lucilia* spp. (Table I).

Over the 2 yr in which we observed *L. silvarum* infestations, 7 amphibians (1 wood frog, 1 Canadian toad, and 5 boreal toads) were found with 2 or more separate lesions; this is the first report of multiple infestation sites by *L. silvarum* on wood frogs or Canadian toads, though James and Maslin (1947) encountered a boreal toad with 3 lesions. In our study, we found lesions most frequently on the hind limbs, back, and flanks of amphibians.

We only documented myiasis by the green blowfly in wild anuran populations in 1998 and 1999, despite trapping in the same study areas using the same methods in 1996 and 1997. Since abundance of *L. silvarum* adults has peaked in July in all population surveys done to date (Dicke and Eastwood, 1952; Schoof et al., 1956; Judd, 1956, 1958), and this period coincided with amphibian trapping periods in our study all 4 yr, it is probable that infestation of amphibians by *L. silvarum* was low enough to be undetected in 1996 and 1997. In northern Alberta, this parasite may typically occur at low levels in wild amphibian populations; prior to this report there has only been one other record of *L. silvarum* myiasis in amphibians in Alberta. Roberts (1998) observed myiasis in 1 wood frog and 3 boreal chorus frogs in 1991 at a site in the aspen parkland of Alberta approximately 350 km south of ours. Interestingly, the years in which we documented *L. silvarum* parasitism (1998 and 1999) and Roberts found parasitized amphibians in southern Alberta (1991) were all relatively dry (Environment Canada, 2004). Roberts (1998) observed that many of the maggots infesting the parasitized wood frog he found exited the wound and drowned when the animal entered water. Perhaps in wet years, when more ephemeral ponds are located across the landscape, amphibians are able to reach water and drown many or all of the *L. silvarum* larvae infesting their bodies.

Recent amphibian surveys involving over 2,000 animals (2001–2006) have not found any parasitism, despite the fact that all captured animals were examined for signs of infestation, suggesting that the incidence of this parasite is very low in Alberta (B. R. E. and C. A. P., unpubl. obs.; C. Browne, pers. comm.), or that infested individuals are killed and consumed so rapidly that they are rarely encountered (Bolek and Janovy, 2004). In the past, *L. silvarum* adults have been collected in Alberta at Lethbridge, Dinosaur Provincial Park, Edmonton, Athabasca, and as far north as the Peace River area (Strickland, 1938), but no information is available on its present distribution in the province. Elsewhere, this species has been reported infesting a live rat (*Rattus norvegicus*) in Wisconsin (Dodge, 1952), a duckling carcass in California (Brothers, 1970), and a human corpse in Colorado (Adair and Kondratieff, 2006); no infestations of nonamphibian species by *L. silvarum* are known from Alberta.

Prevalence of parasitism was higher in species of *Bufo* than in *Rana* or *Pseudacris* species in both years of this study (Table II). Bolek and Janovy (2004) found that *B. americanus* parasitized by *L. silvarum* survived significantly longer than parasitized *R. sylvatica*, though this may have been related to the larger size of the toad hosts. If infestation rates are similar for small and large anurans, but small animals die or are consumed more quickly, smaller infested animals are less likely to be detected. It is also possible that toads, because they are more terrestrial than wood or chorus frogs, are more readily accessible to adult flies and, therefore, acquire more parasites.

Parasitism of amphibians by *L. silvarum* is geographically widespread, with records from across North America (Table I), but occurs at low levels in most years. Here, we have documented: (1) the most northerly occurrence of this parasitism; and what we believe to be (2) both the first report of *L. silvarum* infestation of a boreal toad; and (3) the first record of a *Lucilia* sp. infestation of a Canadian toad (presumed to be *L. silvarum*). We have described what we believe are the first cases of amphibians surviving infestation by this blowfly species. Our data add to the limited knowledge concerning this rarely recorded interaction between amphibians and dipteran parasites.

We thank Kent Kristensen, Jeff Adamyk, Chad Grekul, and Gil Vinkoor for their hard work in the field. Thanks also to Wayne Roberts of the University of Alberta Museum of Zoology for information on fly infestations he observed in amphibians, and to Connie Browne for information on the prevalence of parasitism in her amphibian samples. Comments by two anonymous reviewers improved the manuscript. Funding was provided by a Natural Sciences and Engineering Research Council of Canada (NSERC) Collaborative Special Projects grant and University–Industry Cooperative Research and Development program for the TROLS program. Additional support was provided by Alberta Environmental Protection, Alberta–Pacific Forest Industries, Manning–Diversified Forest Products Research Trust, Syncrude (Canada) Ltd., Weyerhaeuser (Alberta) Ltd., Canadian Circumpolar Institute at the University of Alberta, Alberta Sport, Recreation, Parks and Wildlife Foundation, Challenge Grants in Biodiversity Program (Department of Biological Sciences, University of Alberta, and the Alberta Conservation Association), Ainsworth Lumber, and C&S Resources. Support during manuscript preparation was provided by an NSERC Discovery Grant to C. A. P. Paszkowski.

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