BLOOD PARASITES OF AMPHIBIANS FROM ALGONQUIN PARK, ONTARIO

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ABSTRACT: During a 5 wk period beginning May 25, 1983, 329 amphibians, which included specimens of *Rana catesbeiana* Shaw, *Rana clamitans* Latreille, *Rana septentrionalis* Baird, *Rana sylvatica* LeConte, *Hyla crucifer* Wied, *Bufo americanus* Holbrook, and *Plethodon cinereus* Green, from Lake Sasajewun, Algonquin Park, Ontario, Canada were examined for blood parasites. The prevalences of species of *Trypanosoma*, *Haemogregarina*, *Lankesterella*, *Babesiasoma*, and *Thrombocytozoons* in these amphibians were determined. Two species of microfilaria (probably *Foleyella* spp.) and two intraerythrocytic forms, inclusions of an icosahedral cytoplasmic DNA virus (ICDV) and groups of rickettsial organisms, were also observed. The following are new host records: *Trypanosoma ranarum* (Lankester, 1871) in *B. americanus*; *Trypanosoma ranarum* (Lankester, 1871) in *R. sylvatica*; *Trypanosoma pipientis* Diamond, 1950, *Babesiasoma stableri* Schmittner and McGhee, 1961 and *Thrombocytozoons ranarum* Tchacarof, 1963 in *R. septentrionalis*. The aquatic frogs generally showed a much higher prevalence of infection with blood parasites than the terrestrial frogs, toads and salamanders, which is suggestive of an aquatic vector. The leech *Batracobdella picta* Verrill, 1872, which was found on many of the aquatic frogs, is the most likely vector in the study area. Also, an increasing prevalence of parasites was noted with increasing sizes (ages) of *Rana clamitans* and *R. catesbeiana* suggesting that longer exposure to water makes these species more likely to acquire blood parasites. The presence of *Trypanosoma ranarum* in *B. americanus* appeared to coincide with their attainment of sexual maturity.

INTRODUCTION

In their aquatic and terrestrial habitats, amphibians are exposed to a variety of hematophagous vectors and are consequently in an ideal position to acquire blood parasites. Certain of these parasites from eastern Canada have been described previously (Fantham et al., 1942; Woo, 1969), but in the latter reports neither their prevalence among different host species from one locality nor the relationship between prevalence and host size was considered. In the present study the prevalence of a wide variety of blood parasites of several amphibian species from a sphagnum bog and adjacent forest was determined and examined in relation to the biology of their hosts and potential vectors.

MATERIALS AND METHODS

During the last week of May and throughout June of 1983, 75 bullfrogs (*Rana catesbeiana*

Shaw), 57 green frogs (*Rana clamitans* Latreille), 75 mink frogs (*Rana septentrionalis* Baird), 57 wood frogs (*Rana sylvatica* LeConte), and 10 spring peepers (*Hyla crucifer* Wied) were collected in a sphagnum bog (approximately 50 m by 125 m) on the southwest shore of Lake Sasajewun, Algonquin Provincial Park, Ontario (lat. 45°35′N, long. 78°30′W). Fifty-one specimens of American toads (*Bufo americanus* Holbrook) and four red-backed salamanders (*Plethodon cinereus* Green) were captured in the forest adjacent to Lake Sasajewun during the same period.

The animals were examined for ectoparasites, and snout to vent lengths were recorded. They were marked by toe clipping to ensure that specimens were not re-examined if recaptured. Blood films were prepared by removing the tip of one toe from each specimen (except for the red-backed salamanders from which blood was obtained by removing the tip of the tail) and smearing the blood from the cut face on a slide. The blood films were fixed in methanol, air dried, and stained 8–10 min with Giemsa's stain (1:5 in phosphate-buffered water, pH 7.2). Each film was scanned at low power for several minutes for larger blood parasites such as trypanosomes and microfilariae and then examined for at least 5 min with the oil im-

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mersion 100× objective for intracellular parasites. The parasites were recorded for each animal and measured using an ocular micrometer. All measurements are in μm and are given as a mean followed by the range and the sample size. The photomicrographs were taken on Kodak Panatomic X film in a Zeiss Universal 1 photomicroscope.

Linear regression analyses were performed on raw data concerning the number of species of blood parasites per host versus the size of the host for *R. catesbeiana* and *R. clamitans* because these species show considerable growth after transformation. Significance of the resulting models was tested using one-tailed t-tests and the coefficient of correlation, r, was calculated for each.

Voucher specimens (blood films) containing all the parasites described have been submitted to the National Museum of Canada Invertebrate Collection, Ottawa, Ontario and assigned accession numbers NMCICP1984-0255 through 0265.

RESULTS

An unexpectedly wide variety of parasites was observed in the blood of the amphibians. The most frequently observed parasites were three species of *Trypanosoma*. *Trypanosoma rotatorium* (Mayer, 1843) Laveran and Mesnil, 1901 measured 67.4 (57.8–78.2) by 30.8 (23.8–39.1) (n = 15) (Fig. 1) and was found in the aquatic species of *Rana*. The smaller *Trypanosoma pipientis* Diamond, 1950 which measured 39.2 (36.1–44.3) by 2.6 (1.6–4.1) with a free flagellum measuring 19.4 (16.4–23.0) (n = 10) (Fig. 2) was seen in only two specimens of *Rana septentrionalis*. The stouter and more elongate *Trypanosoma ranarum* (Lankester, 1871) Danilewsky, 1885 measuring 54.6 (32.8–65.6) by 12.1 (9.8–14.8) with a free flagellum measuring 13.6 (4.1–16.4) (n = 15) (Fig. 3) was found in the blood of the larger *Bufo americanus* and in all *Rana* species except *R. septentrionalis*.

Haemogregarine gametocytes were seen in the erythrocytes of some specimens of each of the species of *Rana*. On the basis of their dimensions, staining properties and effect on their host cell and its nucleus, there appeared to be two types. The most frequent type encountered had dark-staining gametocytes which measured 21.8 (20.5–23.0) by 4.6 (4.1–4.9) (n = 15) (Fig. 4) and displaced the host cell nucleus laterally. The second haemogregarine had slightly larger, paler-staining gametocytes which tended to enlarge the host cell and induce its nucleus to become hypertrophied and fragmented (Fig. 5). These latter gametocytes measured 23.6 (21.3–25.4) by 7.2 (5.7–8.2) (n = 15).

Sporozoites of *Lankesterella minima* (Chaussat, 1850) Noller, 1912 were seen occasionally in all species of *Rana* except *R. sylvatica*. The parasites, which occurred exclusively in erythrocytes, measured 12.7 (11.5–14.8) by 2.1 (1.6–3.2) (n = 10), and displayed often a characteristic bulge on their concave surface adjacent to the nucleus (Fig. 6).

A dactylosomid parasite, *Babesiasoma stableri* Schmittner and McGhee, 1961, with an intraerythrocytic cruciform schizont (Fig. 7) was seen often in *Rana septentrionalis* and *R. catesbeiana*. Trophozoites measured 5.9 (4.9–6.6) by 3.6 (2.1–4.1) (n = 10), and large ovoid gametocytes (*sensu* Jakowska and Nigrelli, 1956, and Schmittner and McGhee, 1961) were 9.3 (8.2–12.3) by 3.7 (3.3–4.1) (n = 15).

An unusual parasite, *Thrombocytozoons ranarum* Tchacarof, 1963, was observed in thrombocytes of three of the 75 specimens of *R. septentrionalis*. The elongate parasites laid within a clearly defined vacuole in the thrombocyte cytoplasm, were stained densely and contained two or more pale, spherical inclusions (Fig. 8). The parasites measured 9.5 (7.8–11.5) by 2.6 (1.6–3.3) (n = 31).

Small circular, red-staining inclusions of an icosahedral cytoplasmic DNA virus (ICDV) (Fig. 9) which measured 2.3 (1.8–2.9) (n = 10) in diameter were found in abnormal-appearing erythrocytes of 12 *Rana catesbeiana* and one *R. septentrionalis*. The intensity of infection in some of these frogs was remarkable with up to 90% of the erythrocytes being infected.
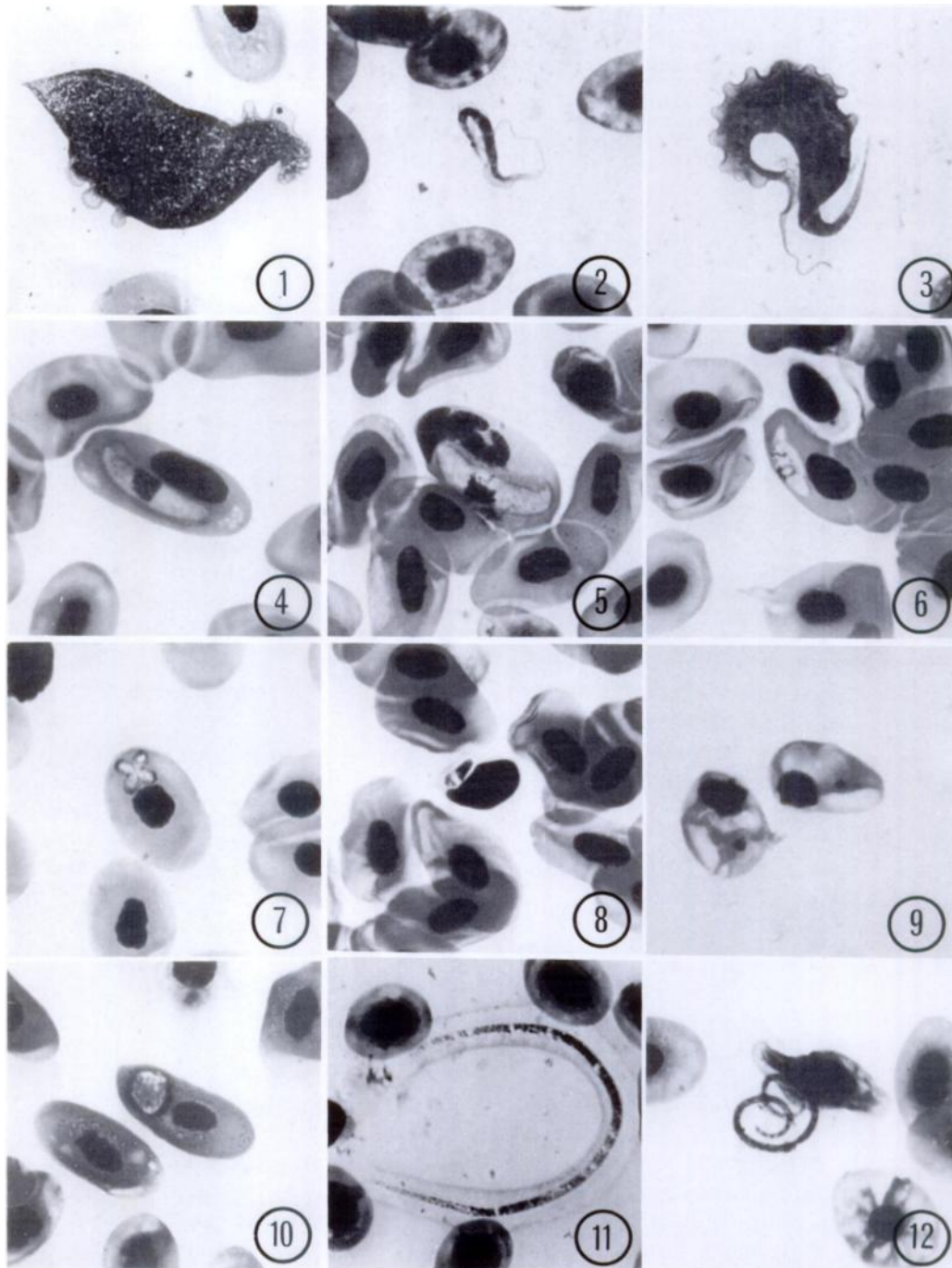
FIGURES 1–12.   Photomicrographs of Giemsa-stained blood parasites of amphibians from the Lake Sasajewun area, Ontario, Canada. All figures ×870. 1. *Trypanosoma rotatorium*. 2. *Trypanosoma pipientis*. 3. *Trypanosoma ranarum*. 4, 5. *Haemogregarina* sp. 6. *Lankesterella minima*. 7. *Babesiasoma stableri*. 8. *Thrombocytozoons ranarum*. 9. Intraerythrocytic icosahedral cytoplasmic DNA virus. 10. Intraerythrocytic inclusions containing slender rickettsia-like prokaryotes. 11, 12. Microfilariae of *Foleyella* spp.

TABLE 1.   Prevalences of blood parasites in Amphibia from Lake Sasajewun, Ontario.

Host species	n	T.r.[a]	T.rn.[b]	T.p.[c]	H[d]	L[e]	B[f]	Th[g]	I[h]	R[i]	M[j]
Aquatic species											
Rana catesbeiana bullfrog	75	52.0	4.0	0.0	30.6	42.6	16.0	0.0	16.0	0.0	0.0
Rana clamitans green frog	57	43.9	10.5	0.0	49.1	14.0	0.0	0.0	0.0	3.5	1.8
Rana septentrionalis mink frog	75	25.3	0.0	2.7	8.0	20.0	18.6	4.0	1.3	0.0	1.3
Terrestrial species											
Rana sylvatica wood frog	57	0.0	3.5	0.0	1.8	0.0	0.0	0.0	0.0	0.0	1.8
Bufo americanus American toad	51	0.0	15.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	2.0
Hyla crucifer spring peeper	10	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Plethodon cinereus red-backed salamander	4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

[a] *Trypanosoma rotatorium.*
[b] *Trypanosoma ranarum.*
[c] *Trypanosoma pipientis.*
[d] *Haemogregarina* sp.
[e] *Lankesterella minima.*

[f] *Babesiasoma stableri.*
[g] *Thrombocytozoons ranarum.*
[h] Icosahedral cytoplasmic DNA virus (ICDV).
[i] Rickettsial inclusions.
[j] Microfilariae.

Larger, pale-staining spherical inclusions, containing numerous rickettsia-like organisms, surrounded by a narrow dark-staining zone, were seen in the erythrocytes of two *Rana clamitans* (Fig. 10). These inclusions measured 7.1 (4.9–10.7) (n = 10).

Microfilariae were rarely seen. One specimen in *Bufo americanus* measured 102.0 by 1.6 (Fig. 11). A second smaller species observed in one *R. clamitans*, one *R. sylvatica* and one *R. septentrionalis* measured 73.8 (65.6–82.0) by 1.2 (n = 4) (Fig. 12).

The only ectoparasites seen on the Amphibia were leeches, almost exclusively *Batracobdella picta* Verrill. These were found frequently on both tadpoles and adults of *R. clamitans*, *R. catesbeiana* and *R. septentrionalis*.

The prevalence of the blood parasites in each of the host species examined is summarized in Table 1. Blood parasites in the aquatic amphibians (*R. catesbeiana,*

R. clamitans and *R. septentrionalis*) had a much higher prevalence than in the more terrestrial species (*H. crucifer*, *B. americanus*, *R. sylvatica* and *P. cinereus*).

The relationship between the average number of species of blood parasites found in *R. clamitans* and *R. catesbeiana*, and the snout to vent lengths (ages) of the frogs is illustrated in Figures 13 and 14, respectively. Although the raw data were used in the regression analyses, only the average of the number of blood parasites per host for each size range and the regression equation were plotted. An increasing number of species of blood parasites was found with increasing sizes (ages) of these frogs. In both *R. clamitans* and *R. catesbeiana* a significant number of infections was discovered in newly transformed or transforming specimens indicating that infections may be acquired as tadpoles from an aquatic hematophagous vector. The relationship between the prevalence of *Trypanosoma ranarum* and the snout
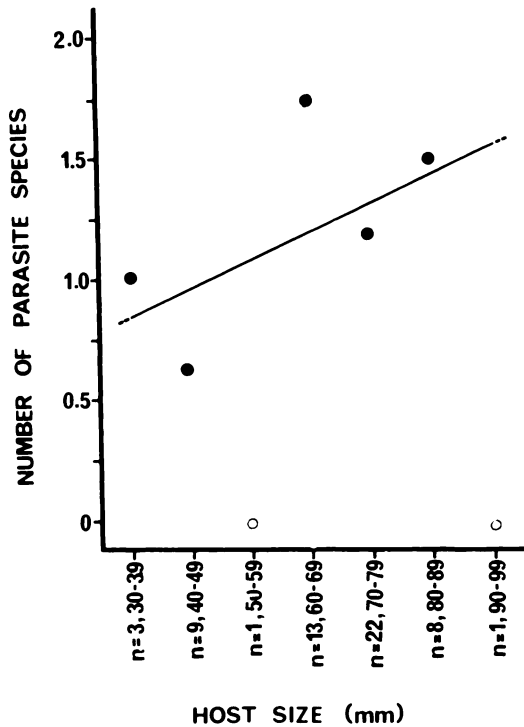
FIGURE 13. The relationship between the average number of blood parasite species per host and the host size (sample size per range, snout to vent length range in mm) is shown for *Rana clamitans*. Solid circles—averages of several specimens; open circles—single specimens (note sample size (n) for each size range). The regression equation is Y = 0.0122X + 0.4155, t = 1.7, df = 55, r = 0.2240.
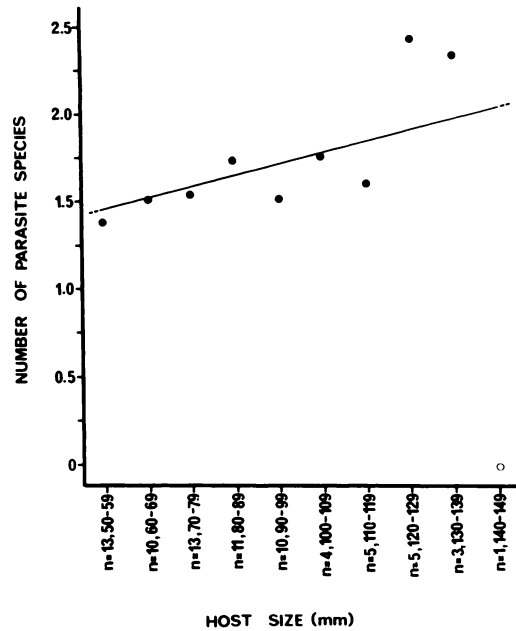

FIGURE 14. The relationship between the average number of blood parasite species per host and the host size (sample size per range, snout to vent length range in mm) is shown for *Rana catesbeiana*. Solid circles—averages of several specimens; open circles—single specimens (note sample size (n) for each size range). The regression equation is Y = 0.078X + 0.9812, t = 1.29, df = 73, r = 0.1472.

to vent lengths of specimens of *Bufo americanus* is indicated in Figure 15. The trypanosomes were seen only in toads with a snout to vent length greater than 50 mm.

DISCUSSION

The life history and habitat of the various hosts will obviously affect their availability to potential vectors of blood parasites. *Rana catesbeiana* spends at least 2 yr as a tadpole before metamorphosing into its adult form. *Rana clamitans* spends 1 yr as a tadpole. Unlike *R. catesbeiana* and *R. clamitans*, the remaining frogs and toads breed in the spring, spend a part of the summer as a tadpole and metamor-

phose into immature adults before the fall. This shorter tadpole phase probably provides fewer opportunities to acquire parasites from an aquatic vector than with *R. catesbeiana* or *R. clamitans*. Like *R. catesbeiana*, *R. clamitans* and *R. septentrionalis* seldom leave water and all three species are considered "aquatic." In contrast, *B. americanus*, *R. sylvatica* and *H. crucifer* leave the water shortly after metamorphosing and return briefly for mating each spring. *Plethodon cinereus* is unusual for an amphibian because it never enters the water. Instead, breeding occurs on land during the fall and spring, and the eggs are attached to the ceiling of a cavity (commonly under a decaying log) by the female. Metamorphosis occurs in the eggs which hatch in August or early
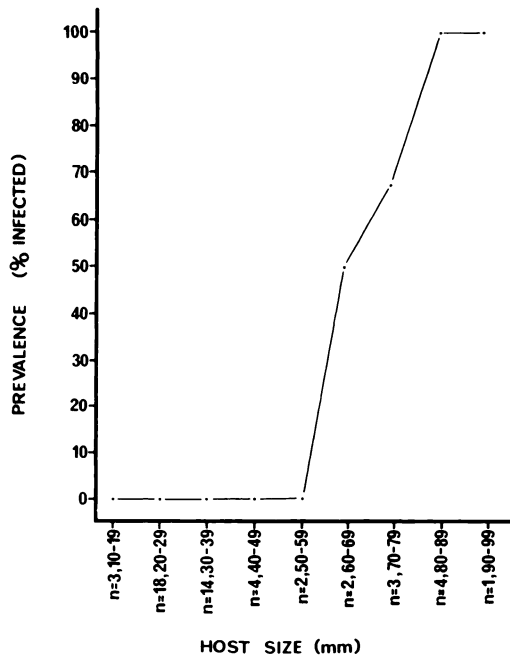
FIGURE 15.   The relationship between the prevalences of *Trypanosoma ranarum* infections and the size of *Bufo americanus* (sample size per range, snout to vent length range in mm). Note the large increase in prevalence in specimens larger than 50 mm in length which have attained sexual maturity.

September. The young remain with the mother for a few weeks before dispersing. Thus, *B. americanus, R. sylvatica, H. crucifer* and *P. cinereus* are considered "terrestrial" (Logier, 1952; Martof et al., 1980).

Trypanosoma rotatorium, the most frequently observed parasite in this study, was limited to the aquatic species of *Rana*. This distribution may reflect a strictly aquatic vector, such as a leech, which would preclude infections of terrestrial amphibians with this parasite. *Trypanosoma pipientis* had not previously been described from *R. septentrionalis*; however, it had been reported in Ontario from *Rana pipiens* (Woo, 1969). *Trypanosoma ranarum* showed the broadest host range with both aquatic and terrestrial hosts. *Trypanosoma ranarum* was originally described from European toads but has been reported from *R. pipiens, R. clamitans* and *R. catesbeiana* in Ontario (Woo, 1969). Data from the present study extend the host range of *T. ranarum* to *R. sylvatica* and *B. americanus*.

Although *Trypanosoma ranarum* has not been described previously from toads in North America, Werner and Walewski (1976) recorded a trypanosome from toads in Michigan which they believed to be *T. bufophlebotomi* Ayala, 1970. Their description and illustration, however, indicated that it was probably *T. ranarum*.

The vector for the trypanosomes in our study area is probably the leech *Batracobdella picta* Verrill, 1872 which seems to feed exclusively on amphibians but is not otherwise host-specific (Sawyer, 1972). Numerous studies have shown that leeches can act as vectors for trypanosomes of amphibians (Brumpt, 1906; Franca, 1915; Barrow, 1953; Diamond, 1958). Although *T. rotatorium* was shown to undergo development and multiplication in a mosquito, *Culex territans* Walker, which feeds primarily on amphibians and is found in the Lake Sasajewun area (Desser et al., 1973), later work by Desser et al. (1975) indicated that this mosquito is not a suitable vector of *T. rotatorium*.

The presence of *T. ranarum* in both aquatic and terrestrial hosts would seem to indicate a different vector from that of *T. rotatorium*. The observed host range could be expected if *T. ranarum* had a terrestrial vector, such as *C. territans*. Photomicrographs taken by Desser et al. (1973) indicated that although only *T. rotatorium* was described, both *T. rotatorium* and *T. ranarum* were present in the blood of the amphibians used for the feeding of *C. territans*. Unfortunately, Desser et al. (1973) did not indicate whether *T. ranarum* was also present in their subsequent transmission study. Therefore one cannot exclude the possibility for transmission of *T. ranarum* by *C. territans*.

Despite our impression of two types of haemogregarine gametocytes in this study,

there was some overlap in their general appearance and dimensions and both "types" frequently occurred in the same animal. Also, the smaller gametocytes were sometimes seen in erythrocytes with a fragmented nucleus.

About a dozen species of *Haemogregarina* have been named from frogs in Africa, Asia and Europe. In North America, with the exception of *Haemogregarina catesbiana* Stebbins, 1904 (from *R. catesbeiana*), *H. boylii* Lehmann, 1959 (from *Rana boyli* Baird, 1854) and *H. aurorae* Lehmann, 1960 (from *Rana aurora*), most haemogregarines from frogs have not been assigned specific names. It is apparent from the photomicrographs of Stebbins (1904) that his description of *H. catesbiana* was based upon a mixture of sporozoites of *Lankesterella* sp., pale-staining spherical intraerythrocytic inclusions similar to those in the present study, and probably immature stages of a species of *Haemogregarina*.

Levine and Nye (1977) found gametocytes of a *Haemogregarina* species in *R. pipiens* in the United States which they claimed corresponded in all respects to *H. magna* (Grassi and Feletti, 1891) Labbé, 1899. Although the haemogregarine(s) in this study are also somewhat similar to *H. magna*, we are reluctant to assign a specific designation. Perhaps the taxonomy of haemogregarines of amphibians should not be further complicated by descriptions of new species until experimental infections in laboratory-reared frogs have been achieved.

Only two species of *Lankesterella* have been described, *L. hylae* (Cleland and Johnston, 1910) from the Australian green tree frog, *Hyla caerulea* White, 1931, and *L. minima* (Chaussat, 1850) Nöller, 1912 from *Rana esculenta* Linnaeus, 1758 in Europe. The dimensions of the sporozoites of the species of *Lankesterella* described herein were similar to those of *L. minima* which has been reported also from *R. pipiens* in the United States by Levine and

Nye (1977). Stehbens (1966) suggested that because *Lankesterella hylae* is found in an arboreal frog, an insect such as a mosquito probably serves as the vector. In contrast, the parasite in our study was found in aquatic frogs and not in terrestrial hosts, suggestive of an aquatic vector.

The dactylosomid parasite encountered frequently in *Rana catesbeiana* and *R. septentrionalis* is *Babesiasoma stableri*. Schmittner and McGhee (1961) found *B. stableri* in *R. pipiens pipiens* in the U.S. and were able to experimentally infect *R. catesbeiana*, *R. pipiens sphenocephala* Cope, 1889, *Bufo terrestris* Bonnaterre, 1789, *B. americanus*, and *B. woodhousei* Girard, 1854 by intraperitoneal injection of heparinized blood from infected animals. The present report of *B. stableri* from *R. septentrionalis* extends its host range in the Ranidae and its geographic distribution. The genus *Dactylosoma*, Labbé, 1894, which with the genus *Babesiasoma* Jakowska and Nigrelli, 1956 comprises the family Dactylosomidae, has been transferred to the Subclass Coccidia from the Subclass Piroplasmia based on ultrastructural work by Boulard et al. (1982). Ultrastructural observations are required to establish the taxonomic positions of the related species of *Babesiasoma*. The absence of *B. stableri* in all of the *R. clamitans* surveyed was peculiar because these frogs were captured in the same area and during the same time period as *R. catesbeiana* and *R. septentrionalis*. The absence of *B. stableri* in *R. clamitans* may be the result of host specificity, but the ease with which additional hosts could be infected experimentally by Schmittner and McGhee (1961) does not support this. Perhaps the feeding habits of the vector or host may prevent transmission although no such feeding preferences were noted for the presumed aquatic vector, *Batracobdella picta*, by Sawyer (1972).

The intrathrombocytic parasite, *Thrombocytozoons ranarum*, found in *R. septentrionalis* in this study was strikingly sim-

ilar to organisms found in *Rana ridibunda* Pallas, 1771 by Tchacarof (1963) in Bulgaria. The prevalence of infection in the Bulgarian frogs was also low (5.4% vs. 4% in *R. septentrionalis*) as was the intensity of infection. Although uncertain of the nature of the "new parasite," Tchacarof (1963) stated that in its affinity for a leucocyte it was similar to the apicomplexan parasites *Leucocytozoon* spp., and hence named it *Thrombocytozoons ranarum.* Examination of these organisms by electron microscopy has revealed that they are prokaryotic, bacillus-like organisms which are surrounded by a cell wall whose size and ultrastructure resembles that of gram-positive bacteria (Desser and Barta, 1984a).

The small red inclusions found in abnormal erythrocytes of *R. septentrionalis* and *R. catesbeiana* have been observed previously in several species of amphibians. Many authors believed that these inclusions were protozoan parasites and therefore erected various genera, such as *Cytamoeba* Labbé, 1894 and *Toddia* Franca, 1911, to accommodate them (Johnston, 1975). Ultrastructural work on the inclusions has shown that many are not protozoan, but are the result of viral infections. The small dark-staining inclusions are now thought to be the result of infection with icosahedral cytoplasmic DNA viruses (ICDV) (Johnston, 1975; Desser and Barta, 1984b).

Preliminary electron microscopic study of the pale-staining, spherical inclusions in the erythrocytes of two specimens of *Rana clamitans* has revealed the presence of numerous slender, elongate rickettsia-like organisms with gram-negative cell walls (Desser and Barta, 1984b). It is noteworthy that bullfrog erythrocytes containing similar inclusions were described and illustrated by Stebbins in 1904, who mistook them for stages of a haemogregarine.

The microfilariae observed in a single toad and the smaller specimens recorded in the green, wood and mink frogs, are probably species of *Foleyella*, a relatively common filarial worm of amphibians.

The large number of parasites found in the aquatic *Rana* species and their rarity in the terrestrial Amphibia indicated that the vector(s) for the *Trypanosoma, Haemogregarina, Lankesterella* and *Babesiasoma* species was most likely aquatic. The distribution of ICDV infections may likewise be suggestive of an aquatic vector. Although host specificity could play a role, the most likely cause for the paucity of blood parasites in terrestrial amphibians is the lack of contact with an aquatic vector. The most notable example is the absence of *Babesiasoma stableri* infections in *Bufo americanus* in an area where numerous infected amphibians reside and a potential vector, *Batracobdella picta*, is present. Schmittner and McGhee (1961) were able to infect *Bufo americanus* by intraperitoneal inoculation of blood containing *Babesiasoma stableri*, but failed to detect the parasite in wild toads. The short breeding period of *Bufo americanus* described by Licht (1976) does not appear to allow effective transmission and establishment of *B. stableri* in the toad population.

The appearance of *Trypanosoma ranarum* infections in specimens of *Bufo americanus* with a snout to vent length above 50 mm (see Fig. 15) serves as another example of host habitat affecting the level of parasitemia. Licht (1976) found that the minimum snout to vent length for mature, breeding *Bufo americanus* in Michigan was approximately 60 mm for males and about 65 mm for females. If the minimum snout to vent length of mature *B. americanus* decreases with increasing latitude, as was found by Schueler (1975) for *R. septentrionalis*, then the expected minimum snout to vent lengths for mature *B. americanus* in this study area could be less than the above figures. The observed increase in prevalence of the trypanosomes may be related to the maturation and breeding of *B.*

americanus. This phenomenon may result from different mechanisms. The vast majority of the toad tadpoles may not be infected before they metamorphose and could subsequently acquire the parasite when they return to the water to breed. This appears unlikely because the probable vector, *B. picta*, is known to feed on *B. americanus* tadpoles (Sawyer, 1972). Alternatively, most tadpoles may become infected before transformation but fail to exhibit detectable peripheral parasitemias until they reach sexual maturity. Thus the trypanosomes would be available to the aquatic vector during the toads' breeding season. Possibly a terrestrial vector, such as *C. territans*, is involved and may feed more readily on mature *B. americanus* during breeding.

The length of time the host is in contact with a potential vector appears to affect the size of the parasite burden. Both *R. catesbeiana* and *R. clamitans* are constantly in contact with water and their parasite load increases with their size. Neither regression equation has a high enough coefficient of regression to be considered a useful predictive model because the variance in the number of blood parasite species per host in each size range is too large. However, the relationship between the host size and the variety of blood parasites is significant as shown by the *t*-test value for each equation. This suggests that longer exposure to the vector is more likely to result in the acquisition of more species of blood parasites. The fact that newly transformed frogs of *Rana* species in this study exhibited significant parasitemias would seem to indicate that parasites are acquired in the tadpole stage as well as in the transformed frogs.

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BOOK REVIEW . . .

Wildlife Disease Review, B. Zimmerman-Haynes and E. A. Edwards, eds. Western Wildlife Laboratories, Inc., 1322 Webster Avenue, Fort Collins, Colorado 80524, USA. 1983. $195.00 (US) in USA and Canada; $250.00 (US) outside USA and Canada.

This is a monthly annotated index to the recent world literature on diseases of captive and free-ranging wildlife. The citations are arranged by major taxonomic groupings of hosts. The preface of the 1983 volume (Volume I) states that "Wildlife Disease Review is a specialized publication designed to provide current, updated literature to veterinarians, wildlife biologists, animal behaviorists, curators, administrators, researchers and students requiring access to the world literature of wildlife diseases." Each entry includes title, author(s), year of publication, journal, volume (number), page(s) and an abstract. Tab divisions for Mammals, Birds, Fish and Reptiles are provided so that new pages can be added conveniently each month to the loose-leaf notebook which is supplied. Each month more than 6,000 international scientific journals are searched for articles concerning wildlife diseases. English abstracts on articles in other languages are given when available. There are four alphabetical indices: (1) a subject index (diseases, etiologic agents, common names of hosts, etc.); (2) a geographic index (countries, regions, areas, states, provinces, etc.); (3) a taxonomic index (scientific names of host species); and (4) an author index. Pages are not numbered, but each citation is assigned a number and they are indexed to those numbers. The format is attractively done on tan paper. This new reference index should prove valuable to anyone interested in keeping up with the literature on wildlife diseases.

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