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NANOPHYETUS SALMINCOLA

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Of 149 wild animals from two western Oregon river drainage areas and other localities in which "salmon poisoning" disease (SPD) is enzootic, including ten mammalian and two avian species, 79 harbored Nanophyetus salmincola, the SPD vector. The shorttail weasel, Mustela erminea; river otter, Lutra canadensis; spotted skunk, Spilogale putorius; Norway rat, Rattus norvegicus; domestic cat, Felis domestica; great blue heron, Ardea herodias; and hooded merganser; Lophodytes cucullatus, are natural hosts not previously reported. Most heavily infected were the raccoon, Procyon lotor, and spotted skunk which apparently are the principal natural definitive hosts. An opossum, Didelphis marsupialis, was infected experimentally for the first time. Six domestic mallard ducks, Anas platyrhynchos, 12 white Leghorn chickens, Gallus gallus, and two mountain voles, Microtus montanus, were refractory to laboratory infection.

DEFINITIVE HOSTS OF THE TREMATODE VECTOR,
NANOPHYETUS SALMINCOLA

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DEFINITIVE HOSTS OF THE TREMATODE VECTOR,
NANOPHYETUS SALMINCOLA¹

INTRODUCTION AND LITERATURE REVIEW

Previous workers have reported natural infections in canids with the "salmon poisoning" trematode, Nanophyetus salmincola. However, Donham, Simms, and Miller (1926) regarded the dog as an accidental host. Philip (1955) suggested that the high incidence of N. salmincola in the snail and fish intermediate hosts could not be explained by infections in canids because "salmon poisoning" disease (SPD) is usually fatal for these animals and they are scarce in the enzootic area. Simms, McCapes, and Muth (1932) stated:

The flukes which transmit the disease to dogs have probably originally come from raccoons, as these are the only carnivores which seem prevalent enough in the salmon poisoning areas to infect the snails of the stream.

Bennington and Pratt (1960) stated:

The most common natural definitive host, has not been determined, but the large numbers of infected snails suggests a very common source of infection, probably raccoon or mink, which eat fish and frequently defecate in or near water.

¹ A portion of this thesis has been previously published according to the following reference. Schlegel, M. W., S. E. Knapp and R. E. Millemann. 1968 "Salmon Poisoning Disease. V. Definitive Hosts of the Trematode Vector, Nanophyetus salmincola. Journal of Parasitology 54:770-774.

Natural infections with N. salmincola in animals other than canids have been reported by Cram (1926) and Simms et al. (1931b). However, there has been no systematic survey of animals in the enzootic area to obtain information concerning the incidence, intensity, and seasonal aspects of natural infections. The present study was undertaken to provide such information.

MATERIALS AND METHODS

Natural Infections

Two study areas were established in western Oregon, one in the Alsea River drainage basin in Benton and Lincoln counties, the other in the Siletz River drainage basin in Lincoln County. Animals were trapped in the former area from September 1965 through June 1966, and in the latter area from October 1966 through June 1967. Included in the survey were the following animals from other localities within the SPD enzootic area: eight raccoons, Procyon lotor; one mink, Mustela vison; one spotted skunk, Spilogale putorius; nine voles, Microtus sp.; four Trowbridge shrews, Sorex trowbridgei; one shrew mole, Neurotrichus gibbsi; one turkey vulture, Cathartes aura, one nutria, Myocastor coypus; one shorttail weasel, Mustela erminea; two opossums, Didelphis marsupialis; two red foxes, Vulpes fulva; one striped skunk, Mephitis mephitis; two coyotes, Canis latrans; one bobcat, Lynx rufus; and one river otter, Lutra canadensis. All were taken in Benton County, except one opossum and both coyotes which were from Clatsop County, the bobcat from Tillamook County, and the river otter from North River, Grays Harbor County, Washington.

Animals were classed either as juveniles less than 12 months of age or as adults. Age classification of raccoons was based on

tooth replacement or wear (Montgomery, 1964), size and curvature of the baculum (Sanderson, 1961), and body weight and measurements.

Trematodes were recovered by washing the contents of the small intestine through a set of Tyler screen sieves the last of which had 100 meshes/inch. The parasites were washed from that sieve into a 1-liter beaker. After they settled, the supernatant fluid was decanted and the beaker refilled with distilled water. Counts of N. salmincola in five 0.1 ml or 1.0 ml samples of the stirred suspension were made and averaged to estimate the number of parasites per animal.

Adult N. salmincola were selected for morphological study from the following: great blue heron (Ardea herodias), bobcat, domestic dog (Canis familiaris), spotted skunk, and raccoon. Each trematode was measured and the number of ova determined. Living parasites were photographed for comparison with mounted specimens.

Fecal samples from the intestine of infected animals except the Norway rat (Rattus norvegicus) and shorttail weasel were examined for N. salmincola ova, by concentration-sedimentation to determine if the parasites were sexually mature.

Laboratory Infections

Metacercariae were given by stomach tube to the following

animals in the numbers and approximate dosages indicated: two mountain voles (Microtus montanus), 5,000 cysts each; twelve 10-week-old white Leghorn chickens, pairs of which received 500, 1,000, 10,000, 25,000, 50,000, or 100,000 metacercariae per bird; six 16-week-old domestic mallard ducks (Anas platyrhynchos), pairs of which were given 1,000, 50,000 or 170,000 cysts per bird; and one opossum given 100,000 metacercariae after daily fecal examinations for 1 week proved negative for ova of N. salmincola.

RESULTS

Natural Infections

Seventy-nine of 149 animals collected, representing ten of the 20 species of mammals and two of the 11 species of birds, were infected with the trematode (Table 1). The shorttail weasel, river otter, spotted skunk, Norway rat, domestic cat, great blue heron, and hooded merganser are natural hosts of N. salmincola not previously reported. Simms et al. (1931b) experimentally infected the domestic cat and the white rat. Donham (1928) reported natural infections in the belted kingfisher but the four belted kingfishers that I examined were negative for N. salmincola.

There were no differences in incidence of infection of the same host species from the two study areas. Also there was no difference between sexes and age of raccoons or between sexes in the spotted skunks. The numbers of other host species were too small to give such information.

As Table 1 shows, raccoons had the highest incidence and intensity of infection with N. salmincola, followed by the spotted skunk. These animals were examined in the largest numbers and contained the most parasites when trapped during December through February. That finding was not surprising because of the coho salmon (Onchorhynchus kisutch) and steelhead trout (Salmo gairdneri)

Table 1. Results of examination of western Oregon birds and mammals for natural infections with Nanophyetus salmincola.

Species	No. infected/ No. examined	No. of parasites	
		Avg per animal	Range
<u>Aix sponsa</u> (wood duck)	0/1		
<u>Ardea herodias</u> (great blue heron)	3/4	14	7-18
<u>Buteo jamaicensis</u> (red-tailed hawk)	0/1		
<u>Cathartes aura</u> (turkey vulture)	0/3		
<u>Corvus brachyrhynchos</u> (common crow)	0/2		
<u>Larus delawarensis</u> (ring-billed gull)	0/1		
<u>Larus philadelphia</u> (Bonaparte's gull)	0/3		
<u>Lophodytes cucullatus</u> (hooded merganser)	2/6		12-24
<u>Megacercyle alcyon</u> (belted kingfisher)	0/4		
<u>Melanitta deglandi</u> (white-winged scoter)	0/1		
<u>Mergus merganser</u> (American merganser)	0/8		
<u>Canis latrans</u> (coyote)	1/3		72,000
<u>Didelphis marsupialis</u> (opossum)	0/2		
<u>Felis domesticus</u> (domestic cat)	1/2		36
<u>Lutra canadensis</u> * (river otter)	2/3		2-4
<u>Lynx rufus</u> (bobcat)	6/8	3,825	200-11,000
<u>Mephitis mephitis</u> (striped skunk)	0/1		
<u>Microtus oregoni</u> and <u>M. townsendi</u> (Oregon and Townsend vole)	0/9		
<u>Mustela erminea</u> (shorttail weasel)	1/2		2
<u>Mustela vison</u> (mink)	2/5		10-450
<u>Myocaster coypus</u> (nutria)	0/1		
<u>Neotoma cinerea</u> (busytail woodrat)	0/1		
<u>Neurotrichus gibbsi</u> (shrew mole)	0/1		
<u>Ondatra zibethica</u> (muskrat)	0/1		
<u>Procyon lotor</u> (raccoon)	42/42	57,571**	8-475,000
<u>Rattus norvegicus</u> (Norway rat)	3/6	24	20-30
<u>Sorex trowbridgei</u> (Trowbridge shrew)	0/4		
<u>Spilogale putorius</u> (spotted skunk)	15/20	2,613**	1-17,500
<u>Ursus americanus</u> (black bear)	0/1		
<u>Vulpes fulva</u> (red fox)	1/3		2,400

*One of the two infected animals came from North River in western Washington.

**For 37 raccoons and 13 spotted skunks.

are most abundant in Oregon coastal streams during the late fall and winter. Hence, the maximal opportunity for definitive hosts to become infected occurs at that time of the year.

Parasites from the raccoon also contained the most ova, the maximum number per worm being 27, followed by 24 for the spotted skunk and 21 for the red fox. In other hosts, the number was: coyote, nine; mink, five; bobcat, domestic cat, and great blue heron, three; shorttail weasel and river otter, two; hooded merganser, one; and Norway rat, zero.

Fecal examinations revealed ova of N. salmincola in the bobcat, domestic cat, coyote, mink, raccoon, red fox, and spotted skunk, but not in the river otter, great blue heron, or hooded merganser.

Trematodes from the great blue heron, hooded merganser, river otter, and bobcat were atypical in body shape. The largest N. salmincola found were from the great blue heron, and the smallest from the spotted skunk (Table 2). The possible influence of host species on parasite morphology is under investigation.

Laboratory Infections

The mountain voles were examined 7 days after exposure, and the chickens and mallard ducks were examined after 14 days but no trematodes were found. Donham (1928) also was unable to infect

experimentally two chickens and one domestic duck. Examination of the opossum 8 days after exposure yielded 19,000 adult N. salmincola. The feces contained N. salmincola ova. The animal showed no signs of SPD.

Table 2. Average measurements of Nanophyetus salmincola.

<u>Species</u>	No. Animals Examined	No. Flukes Measured	Length mm	Width mm
<u>Ardea herodias</u> (great blue heron)	1	7	1.488	0.643
<u>Lynx rufus</u> (bobcat)	4	34	1.015	0.494
<u>Canis familiaris</u> (dog)	3	30	1.157	0.586
<u>Procyon lotor</u> (raccoon)	5	26	0.758	0.433
<u>Spilogale putorius</u> (spotted skunk)	2	8	0.439	0.200

DISCUSSION

This study supports Simms et al. (1932) and Bennington and Pratt (1960), who believed the raccoon or some other fish-eating animal likely to be responsible for the perpetuation of the trematode in nature. Our findings indicate that the raccoon is the principal natural definitive host because: (1) it had the highest incidence and intensity of infection; (2) its trematodes had the largest number of ova; and (3) excluding rodents, the raccoon is probably the commonest mammal in coastal stream areas of western Oregon. In his summary of the literature on population densities of North American small mammals, Mohr (1947) reported that the number of raccoons per acre is greater than that of coyotes, red foxes, and mink. Not included in that summary were population densities for other species of mammals which we found parasitized by N. salmincola. After the raccoon, the spotted skunk appears to be the most important natural definitive host because of high incidence and intensity of infection, the large number of ova per parasite, and the abundance of the animal in the enzootic area.

The river otter, mink, and the fish-eating birds examined were not heavily infected, although fish are always accessible to them. Because most of the mink and otter had been dead two or more days prior to examination, the fragile parasites may have

deteriorated and not been recognized. However, finding parasites with atypical morphology and few eggs in the great blue heron, hooded merganser, and river otter suggests that these animals may not be optimal hosts for N. salmincola. Table 3 lists 23 natural and experimental definitive hosts for our strain of N. salmincola and Table 4 lists 15 for the Siberian strain which Skrjabin and Podjapolskaja (1931) described as a new species, N. schikhobalowi. That name is considered by Witenberg (1932), Gebhardt et al. (1966), and Filimonova (1966) to be a synonym of N. salmincola. Natural human infections have not been reported in the USA but Philip (1958) successfully infected himself experimentally, and N. salmincola is a parasite of natives of eastern Siberia. Natural definitive hosts common to the two strains are the domestic dog, domestic cat, red fox, and mink; common experimental hosts are the domestic dog, domestic cat, and white rat.

It is apparent that N. salmincola has little definitive host specificity, and Gebhardt et al. (1966) reported the same for the second intermediate host. These findings agree with the long-held belief that the restricted geographic range of our strain of N. salmincola is determined by that of the snail host, Oxytrema silicula, whose range coincides with the SPD enzootic area (Simms et al. 1931a). That situation implies that the trematode is highly specific for O. silicula; however, experimental proof for such specificity is still lacking.

Table 3. Definitive hosts for Nanophyetus salmincola in northwestern USA.

Host	Observed infection		Reference
	N*	E	
Man	-	+	Philip, 1958
<u>Ardea herodias</u> (great blue heron)	+	-	Present paper
<u>Lophodytes cucullatus</u> (hooded merganser)	+	-	Present paper
<u>Megaceryle alcyon</u> (belted kingfisher)	+	-	Donham, 1928
<u>Alopex lagopus</u> (arctic fox)	+	+	Simms et al., 1931a, b
<u>Canis familiaris</u> (domestic dog)	+	+	Many authors
<u>Canis latrans</u> (coyote)	+	+	Cram, 1926; Donham and Simms, 1927; present paper
<u>Cavia porcellus</u> (guinea pig)	-	+	Simms et al, 1931b
<u>Didelphis marsupialis</u> (opossum)	-	+	Present paper
<u>Felis domestica</u> (domestic cat)	+	+	Simms et al., 1931b; present paper
<u>Lutra canadensis</u> (river otter)	+	-	Present paper
<u>Lynx rufus</u> (bobcat)	+	+	Cram, 1926; Simms et al., 1931b; present paper
<u>Meriones unguiculatus</u> (jird)	-	+	Nyberg and Knapp (unpublished)
<u>Mesocricetus auratus</u> (golden hamster)	-	+	Philip, 1959; Bennington and Pratt, 1960
<u>Mus musculus</u> (white mouse)	-	+	Philip, 1959
<u>Mustela erminea</u> (shorttail weasel)	+	-	Present paper
<u>Mustela vison</u> (mink)	+	-	Donham et al., 1926; Simms et al., 1931b; Baker, 1950; present paper
<u>Neotoma</u> sp. (woodrat)	-	+	Bennington and Pratt, 1960
<u>Procyon lotor</u> (raccoon)	+	+	Cram, 1926; Simms et al., 1931b; Bennington and Pratt, 1960; present paper
<u>Rattus norvegicus</u> (Norway rat)	+	+	Simms et al., 1931b; present paper
<u>Spilogale putorius</u> (spotted skunk)	+	-	Present paper
<u>Ursus americanus</u> (black bear)	-	+	Simms et al., 1931b
<u>Vulpes fulva</u> (red fox)	+	+	Donham et al., 1926; Simms et al., 1931b; Cordy and Gorham, 1950; present paper

*N = natural and E = experimental. Negative sign indicates no information available or evidence inconclusive because of small sample size.

Table 4. Definitive hosts for Nanophyetus salmincola (= N. schikhobalowi) in eastern Siberia.*

Host	Observed infection		Reference
	N**	E	
Man	+	-	Filimonova, 1963, 1966
<u>Canis familiaris</u> (domestic dog)	+	+	Filimonova, 1963, 1966
<u>Canis lupus</u> (gray wolf)	+	-	Filimonova, 1966
<u>Enhydra lutris</u> (sea otter)	+	-	Filimonova, 1966
<u>Felis domestica</u> (= <u>F. catus</u>) (domestic cat)	+	+	Filimonova, 1963, 1966
<u>Gulo gulo</u> (wolverine)	+	-	Filimonova, 1966
<u>Martes flavigula</u> (yellow-throated Asiatic marten)	+	-	Filimonova, 1966
<u>Meles meles</u> (Old World badger)	+	-	Filimonova, 1966
<u>Mustela sibirica</u> (Kolinsky)	+	+	Filimonova, 1965, 1966
<u>Mustela vison</u> (mink)	+	+	Filimonova, 1965, 1966
<u>Nyctercutes procyonoides</u> (raccoon dog)	+	-	Filimonova, 1966
<u>Rattus norvegicus</u> (Norway rat)	-	+	Filimonova, 1963, 1965
<u>Selenarctos tibetanus</u> (Asiatic black bear)	+	-	Filimonova, 1966
<u>Ursus arctos</u> (Alaskan brown bear)	+	-	Filimonova, 1966
<u>Vulpes vulpes</u> (red fox)	+	-	Filimonova, 1966

*Locality for the sea otter was the Commander Islands.

**See footnote in Table 2.

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