



Contents lists available at ScienceDirect

International Journal for Parasitology: Parasites and Wildlife

journal homepage: www.elsevier.com/locate/ijppaw

Endoparasites of American marten (*Martes americana*): Review of the literature and parasite survey of reintroduced American marten in Michigan



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ARTICLE INFO

Article history:

Received 1 April 2016

Received in revised form

2 July 2016

Accepted 9 July 2016

Keywords:

American marten

Endoparasite

Faecal examination

Michigan

Wildlife

Zoonotic

ABSTRACT

The American marten (*Martes americana*) was reintroduced to both the Upper (UP) and northern Lower Peninsula (NLP) of Michigan during the 20th century. This is the first report of endoparasites of American marten from the NLP. Faeces from live-trapped American marten were examined for the presence of parasitic ova, and blood samples were obtained for haematocrit evaluation. The most prevalent parasites were *Capillaria* and *Alaria* species. Helminth parasites reported in American marten for the first time include *Eucoleus boehmi*, hookworm, and *Hymenolepis* and *Strongyloides* species. This is the first report of shedding of *Sarcocystis* species sporocysts in an American marten and identification of 2 coccidian parasites, *Cystoisospora* and *Eimeria* species. The pathologic and zoonotic potential of each parasite species is discussed, and previous reports of endoparasites of the American marten in North America are reviewed.

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1. Introduction

The American marten (*Martes americana*) is an arboreal meso-carnivore that ranges from the boreal forests of northern North America into coniferous and mixed coniferous/deciduous forests of the northern and northeastern United States, including the Great Lakes region (Clark et al., 1987). The American marten was reintroduced to Michigan's Upper Peninsula (UP) and northern Lower Peninsula (NLP) in the mid-20th century after regional extirpation due to habitat loss and over-harvest (Cooley et al., 2004). Over 200 animals were reintroduced to the UP over the course of several reintroductions. Many fewer animals (n = 36) were reintroduced in the Huron-Manistee National Forest of the NLP and 49 martens were reintroduced to the Pigeon River State Forest of the NLP. The UP population has since grown and currently sustains an annual harvest for fur. In contrast, the species is considered a Regional Forester Sensitive Species and there is no harvest in the NLP (USDA

Forest Service, 2012). Factors hypothesized to be contributing to the differences in population sustainability between the UP and NLP include differences in habitat, genetic diversity, health and others. At the time of the original reintroduction, American marten were not examined for parasitic or infectious diseases (Spriggs, unpublished data). Some parasites are of economic or zoonotic importance and may be introduced with animal translocations. Therefore, reintroduction programs should take into account the presence of parasites which are pathogenic or to which the species of concern is not adapted (Kimber and Kollias, 2000).

A collaborative research effort has begun in order to investigate factors that may be contributing to the difference in sustainability between the UP and NLP populations. The aim of this parasite survey was to describe the presence and prevalence of parasites in the Huron-Manistee National Forest of the NLP and to determine whether there are differences in presence or prevalence of parasites between the NLP and UP. This survey uses fecal examination as a non-invasive method for identifying parasites. Should future translocation of animals from the UP into the NLP be considered for management of the species, a non-invasive and inexpensive method for screening animals would be desirable. Also, American marten are not harvested in the NLP and thus adequate numbers of

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carcasses are not available for examination.

While some information exists regarding the prevalence of endoparasitism in American marten in North America, relatively little information is available for the species in Michigan (Poole et al., 1983; Veine-Smith et al., 2011). This study reviews previous parasite prevalence reports from American marten in North America, presents data from the parasitological examination of live-trapped American marten in Michigan, and identifies a possible association between hookworm infection and anaemia in affected American marten.

2. Materials and methods

American marten ($n = 49$) were sampled from the Manistee National Forest in the NLP ($n = 31$), Hiawatha National Forest in the UP ($n = 13$) and Ottawa National Forest in the UP ($n = 5$). American marten were trapped and immobilized from 2011 to 2015 as described by Desmarchelier et al. (2007) for a concurrent habitat study. Faeces were collected either via fecal loop from the rectum or from the trap. Faeces were stored under refrigeration and examined within 4 days of collection. Nine American marten were recaptured and re-sampled, resulting in a total of 60 faecal samples included in analyses.

Blood was collected from the jugular vein and placed into lithium heparin anticoagulant (BD Microtainer Tubes, Becton Dickinson and Company). We determined haematocrit using microhematocrit capillary tubes (SafeCrit) and the StatSpin VT centrifuge (Iris). Individuals were identified as anaemic if the haematocrit was $<42\%$ based on a report of normal haematocrit in captive, fed American martens of $47 \pm 5\%$ (Nieminen et al., 2007). Other health parameters were collected as part of a complete health assessment (Spriggs, unpublished data). Faecal float and sedimentation examinations were performed at Michigan State University's Diagnostic Center for Population and Animal Health using standard methods (Zajac and Conboy, 2012). Faecal flotation was performed using Sheather's sugar solution (specific gravity 1.25–1.27) alone ($n = 30$) from May 2011–May 2012 or with both Sheather's sugar solution and zinc sulfate solution (specific gravity 1.18, $n = 30$) from June 2012–January 2015 and examined by light microscopy. Fifty-nine of the samples, representing 48 individual animals, were sufficient in quantity for faecal sedimentation procedure. Ova were identified based on morphologic characteristics including size and in accordance with parasitologic references and previous reports. Upon consultation with a wildlife veterinary parasitologist (Gerhold), 2 nematode species (*Syphacia muris* and *Aspicularis* sp.) were presumed to be pass-through from rodent prey and excluded from analyses.

Statistical analysis was performed with JMP Pro 10.0.02 (SAS Institute Inc.). Prevalence was calculated as the number of infected hosts divided by the number of hosts examined. Because some American marten were sampled more than once, all parasite species found in an individual were considered together for prevalence calculations. Differences between locations (UP and NLP) and sexes were examined with a Pearson's χ^2 test with $P < 0.05$ considered significant. Significant differences between presence or absence of anaemia and hookworm infection were also examined with a Pearson's χ^2 test. Parasite species richness by host was calculated as the number of parasite species present per host; species richness by sample was calculated as the number of parasite species present per sample. An unidentified capillarid species was not included in the prevalence or host species richness if one or more species of *Capillaria* was identified in other samples from the same host. Thus, an individual American marten found with unidentified capillarid at the initial exam and *Aonchotheca* sp. at a subsequent exam was considered to be infected with *Aonchotheca* sp. alone when

reporting prevalence. If both samples had unidentified *Capillaria*, then the host was included in the calculation of prevalence of unidentified *Capillaria*. Unidentified *Capillaria* were always included in the sample species richness calculations. Differences between sex and location in species richness were examined with a Wilcoxon rank sum test, with $P < 0.05$ considered significant.

The capture and handling protocol was approved by the University of Tennessee Animal Care and Use Committee (protocol #2180), and American marten live-trapping and sample collection was an authorized tribal activity under the 2007 Inland Consent Decree between the State of Michigan and the Little River Band of Ottawa Indians.

3. Results

Sixty samples from 49 individual American marten (28 males, 21 females) were examined, and results are shown in Table 1. Parasite species richness by host is shown in Table 2. Of 49 individual American marten examined, 91.8% were positive for 1 or more parasites and 69.4% were infected with 2 or more parasites. There was no significant difference in mean species richness by host between sexes or locations (NLP and UP).

Trematode eggs were seen in over half (63.3%) of all hosts. *Alaria* species was suspected or confirmed, depending upon examination method. Trematode egg identification was suspected but not confirmed to be *Alaria* sp. during the early part of the study (May 2011–May 2012) when Sheather's sugar solution alone was used for flotation, as *Alaria* species may be distorted due to the osmotic pressure of sugar solution. Later in the study (June 2012–January 2015), trematode ova found on sedimentation were confirmed to be *Alaria* species when the sample was floated with zinc sulfate solution, in which trematode ova will not be distorted. Once the use of zinc sulfate solution was implemented, no trematode other than *Alaria* species was identified, and the authors concluded that trematode eggs identified in the early samples were most likely *Alaria* species, as suspected. In the UP, 3 samples that were positive on sedimentation for trematode ova were suspected to be *Alaria* species and 7 were confirmed via zinc sulfate flotation; in the NLP, 14 samples positive on sedimentation were suspected to be *Alaria* species and 5 were confirmed via zinc sulfate flotation. *Capillaria* eggs were seen in 79% and 78.1% of samples from the UP and NLP, respectively. *Capillaria* eggs were further identified as *Eucoleus aerophila*, *Eucoleus boehmi*, or *Aonchotheca putorii* based on size and morphologic characteristics. A hookworm egg is shown in Fig. 1. There was no significant difference in prevalence of any of the identified parasites between male and female American marten. Of 9 American marten that were sampled more than once, only one had identical results for each time point.

The mean haematocrit was 45.6 ± 8.1 (range 30–68; $n = 49$). Haematocrit was reported to be $47 \pm 5\%$ in the only other report of American marten haematocrit (Nieminen et al., 2007). Using $<42\%$ as a cut-off, 24.5% of blood samples tested in this report were considered anaemic. American marten infected with hookworms were significantly more likely to be anaemic than non-infected American marten ($P = 0.01$) with an odds ratio of 8.75 (95% confidence interval: 1.4–56.4).

4. Discussion

Parasite species richness per host was similar to that reported by Veine-Smith et al. (2011) for American marten in the UP. We identified more parasite species in the NLP than the UP, but this result may be a function of the larger sample size from the NLP, and there was no significant difference in richness between the 2 locations. Foreyt and Langerquist (1993) found 2 or more parasites in

Table 1

Prevalence of parasite species identified in faecal samples from live-trapped American marten in the Upper Peninsula (UP) and northern Lower Peninsula (NLP), Michigan. Significant differences in parasite prevalence between the locations are indicated by the same superscript ($P < 0.05$).

Parasite	Prevalence UP % (n = 18)	Prevalence NLP % (n = 31)
Protozoa		
Coccidia	27.8	25.8
<i>Sarcocystis</i>	0	3.2
Trematode		
<i>Alaria</i> sp. ^a	55.6	63.3
Cestode		
<i>Hymenolepis</i>	0	3.2
Nematodes		
Capillaria		
<i>Eucoleus aerophila</i>	16.7	3.2
<i>Eucoleus boehmi</i>	0	9.7
<i>Aonchotheca putorii</i>	77.8 ^a	29.0 ^a
Unidentified capillaria	0 ^b	42.0 ^b
<i>Crenosoma vulpis</i>	11.1	0
Hookworm	11.1	16.1
<i>Physaloptera</i>	5.6	3.2
<i>Strongyloides</i>	0	3.2

^a Only 30 marten from the NLP had faecal sample quantities adequate for sedimentation and inclusion in prevalence calculations. Prevalence shown here reflect samples in which *Alaria* species were suspected or confirmed.

^b Statistical comparison was not made between prevalence of unidentified parasites.

Table 2

Mean species richness by host for American marten by location and sex. There were no significant differences in richness between the locations or sexes.

Category	Mean species richness by host (SD)	Range	n
Overall	2.0 (1.1)	0–5	49
UP	2.1 (1.1)	0–4	18
NLP	2.0 (1.2)	0–5	31
Males	2.1 (1.2)	0–5	28
Females	2.0 (1.0)	0–4	21



Fig. 1. Hookworm egg from an American marten (*Martes americana*). There was no significant difference in the prevalence of hookworm eggs in faecal samples from American marten from the Upper Peninsula (n = 18) and Northern Lower Peninsula (n = 31) of Michigan, USA (prevalence 11.1% and 16.1%, respectively). American marten infected with hookworms were significantly more likely to be anaemic than non-infected American marten ($P = 0.01$) with an odds ratio of 8.75 (95% confidence interval: 1.4–56.4).

35% of American marten from Eastern Washington. In another survey from Washington, 9 helminth species were found, and 48.4% of hosts had coinfection of 2 or more parasites, with a maximum of 4 (Hoberg et al., 1990). Overall, parasite prevalences were similar between the UP and NLP in our samples and we conclude that the UP may be suitable source for a translocation of marten to the NLP from the point of view of parasite translocation. Additional screening of both populations, the use of molecular techniques to definitively speciate parasites, and testing of individual martens marked for translocation are recommended.

Parasites in our study were identified to the genus and/or species level as possible by microscopic examination alone. Future research could employ the use of molecular tools such as polymerase chain reaction to provide more specific results. Such techniques were not used in this study because we desired an inexpensive method that could be repeated in any future reintroduction programs that require screening of large numbers of animals. Parasites identified as a concern for reintroduction of North American river otter included *Alaria canis*, *Strongyloides lutrae*, *Crenosoma goblei*, *Capillaria* and coccidia due to potential for pathogenic effects or high prevalence (Kimber and Kollias, 2000). River otter reintroduction programs tested and treated parasitized otters, and similar methods could be considered for reintroduction of American marten (Hoover et al., 1985; Griess, 1987; Serfass et al., 1993). Of 9 American marten that were sampled more than once, only one had identical results at subsequent sampling, indicating either a change in infection status or inconsistent shedding of ova in the remaining 8 martens. Therefore, multiple screening faecal parasitological examinations are warranted. Endoparasites previously reported in North American marten are presented in Table 3.

4.1. Trematodes of American marten in Michigan

Alaria species are flukes found in the small intestine of definitive hosts including felids, canids and mustelids. *Alaria mustelae* is known to infect mink (*Mustela vison*) and short-tailed weasels (*Mustela erminea*), as well as American marten (Veine-Smith et al., 2011). *Alaria taxidae* was identified from 25% (n = 6) of American marten from the District of Mackenzie, Northwest Territories and was identified in Manitoba, Canada, at prevalences ranging from 36 to 73% depending on the area (Holmes, 1963; Poole et al., 1983).

Table 3
Helminth parasites previously reported in American marten.

Genus and species	Location	Prevalence (%)	Infected (n)	Study
Nematodes				
<i>Baylisascaris devosi</i>	Northern Cascades, Washington, USA	21.0	3	Hoberg et al., 1990
	Southern Cascades, Washington, USA	2.0	1	Hoberg et al., 1990
	Manitoba, CA	0.7	1	Poole et al., 1983
<i>Capillaria putorii</i>	Eastern Washington, USA	86.0	36	Foreyt and Langerquist, 1993
	Upper Peninsula, Michigan, USA	47.0	66	Veine-Smith et al., 2011
<i>Crenosoma</i>	Colorado, USA	29.0	18	Olsen, 1952
	Upper Peninsula, Michigan, USA	2.0	3	Veine-Smith et al., 2011
	Ontario, CA	0.2	1	Seville and Addison, 1995
<i>Diocotophyme renale</i>	Ontario, CA	2.0	8	Seville and Addison, 1995
<i>Dracunculus insignis</i>	Ontario, CA	0.2	1	Seville and Addison, 1995
<i>Eucoleus aerophilus</i>	Ontario, CA	4.0	16	Seville and Addison, 1995
<i>Filaroides martis</i>	Upper Peninsula, Michigan, USA	4.0	5	Veine-Smith et al., 2011
	Ontario, CA	8.0	37	Seville and Addison, 1995
<i>Molineus patens</i>	Southern Cascades, Washington, USA	9.0	6	Hoberg et al., 1990
<i>Pearsonema plica</i>	Ontario, CA	6.0	24	Seville and Addison, 1995
<i>Physaloptera</i>	Southern Cascades, Washington, USA	2.0	1	Hoberg et al., 1990
	Upper Peninsula, Michigan, USA	9.0	13	Veine-Smith et al., 2011
<i>Soboliphyme baturini</i>	Northern Alaska, USA	19.0	416	Zarnke et al., 2004
	Southeastern Alaska, USA	55.0	85	Thomas et al., 2008
	Southeastern Alaska, USA	47.0	1430	Zarnke et al., 2004
	Southwestern Alaska, USA	30.0	321	Zarnke et al., 2004
	Northern Cascades, Washington, USA	7.0	1	Hoberg et al., 1990
<i>Trichinella spiralis</i>	Idaho, USA	0.9	1	Erickson, 1946
	British Columbia, CA	61.0	22	Schmitt et al., 1976
	Northern Cascades, Washington, USA	50.0	7	Hoberg et al., 1990
	Southern Cascades, Washington, USA	31.0	20	Hoberg et al., 1990
	Eastern Washington, USA	5.0	2	Foreyt and Langerquist, 1993
	Manitoba, CA	0.7	1	Poole et al., 1983
	Montana, Idaho and Wyoming, USA	8.3	2	Worley et al., 1974
	Ontario, CA	3.4	68	Dick et al., 1986
	Ontario, CA	2.0	8	Seville and Addison, 1995
Quebec, CA	1.8	1	Bourque, 1985	
Trematodes				
<i>Alaria</i>	Duck Mountain, Manitoba, CA	73.0	45	Poole et al., 1983
	Porcupine Mountain, Manitoba, CA	57.0	4	Poole et al., 1983
	Southern Indian Lake, Manitoba, CA	36.0	32	Poole et al., 1983
	Upper Peninsula, Michigan, USA	39.0	54	Veine-Smith et al., 2011
	Northwest Territories, CA	25.0	6	Holmes, 1963
<i>Euryhelmis squamula</i>	Southern Cascades, Washington, USA	6.0	4	Hoberg et al., 1990
Cestodes				
<i>Mesocestoides</i>	Northern Cascades, Washington, USA	21.0	3	Hoberg et al., 1990
	Southern Cascades, Washington, USA	59.0	38	Hoberg et al., 1990
	Eastern Washington, USA	33.0	14	Foreyt and Langerquist, 1993
<i>Taenia martis americana</i>	Northern Cascades, Washington, USA	14.0	2	Hoberg et al., 1990
	Southern Cascades, Washington, USA	16.0	10	Hoberg et al., 1990
	Southern Indian Lake, Manitoba, CA	23.0	16	Poole et al., 1983 ^a
<i>Taenia mustelae</i>	Southern Cascades, Washington, USA	30.0	19	Hoberg et al., 1990
	Duck Mountain, Manitoba, CA	15.0	9	Poole et al., 1983
	Northwest Territories, CA	29.0	7	Holmes, 1963
	Northwest Territories, CA	12.5	3	Holmes, 1963
Protozoa				
<i>Cryptosporidium</i>	Upper Peninsula, Michigan, USA	4.0	5	Veine-Smith et al., 2011
<i>Giardia</i>	Upper Peninsula, Michigan, USA	4.0	5	Veine-Smith et al., 2011
<i>Sarcocystis</i>	Eastern Washington, USA	10.0	4	Foreyt and Langerquist, 1993
<i>Toxoplasma gondii</i>	Ontario, CA	10.8	15	Tizard et al., 1976
	Michigan, USA	58.0	47	Poole et al., 1983
	Manitoba, CA	15.0	9	Poole et al., 1983
	Northwest Territories, CA	29.0	7	Holmes, 1963
	Northwest Territories, CA	12.5	3	Holmes, 1963

^a *Taenia* species (cf. *martis martis*).

A definitive host infected with *Alaria* species in its intestines sheds eggs in its faeces. After 2 weeks in wet soil or water, the eggs hatch, producing a miracidium. The miracidium invades a fresh-water snail, at which point it develops into a cercaria. The cercaria invades a tadpole and develops into a mesocercaria. When the tadpole is ingested by an amphibian, reptile, or rodent, the mesocercariae remain in the tissues of the paratenic host. When a carnivore consumes a paratenic host, the parasite completes its life cycle. The mesocercariae migrate through the stomach, across the

diaphragm and into the lungs where the mesocercariae develops into metacercariae. The metacercariae are able to travel up the trachea and are then swallowed, at which point they develop into adult trematodes in the small intestines. While infection with *Alaria* species typically does not cause disease in the definitive host, there are reports of the mesocercariae causing neurologic disease due to aberrant migration through the central nervous system and of respiratory illness due to migration through the lungs in domestic dogs (Kimber and Kollias, 2000; Kazacos, 2001). Other species of

trematodes have caused disease in North American river otters, but the pathogenicity of *Alaria* in American marten is not known (Kimber and Kollias, 2000).

If a pregnant female rodent or carnivore becomes infected with *Alaria*, the mesocercariae can migrate to the mammary glands and can be transmitted to nursing young (Bowman et al., 2009). In an American marten kit, not yet weaned, in the NLP, histopathology revealed infection with *Alaria* species in the duodenum, confirming the potential for lactogenic transmission of mesocercariae in this mustelid.

In the report by Veine-Smith et al. (2011), faeces from the large intestines of 140 American marten carcasses from the UP were examined for flukes using a sedimentation technique and found a prevalence of 39% (n = 54). The difference in prevalence between that report and ours (55.6%, n = 10) may be due to differences in sample size, methodology or a true difference (Veine-Smith et al., 2011). We recommend the use of both faecal sedimentation and zinc sulfate flotation for identification of trematode eggs in faecal samples obtained from live animals.

4.1.1. Other reported trematodes of American marten in North America

Euryhalmis squamula has been reported in raccoons (*Procyon lotor*), mink, and American marten in Washington and uses amphibian intermediate hosts in this region (Hoberg et al., 1990). American marten from Washington were documented with 6% prevalence from the southern Cascades, confirming that marten are ingesting anuran prey in this area (Hoberg et al., 1990). *E. squamula* has been reported in mink in North America and is a common parasite of the polecat in Europe (Ameel, 1938; Miller and Harkema, 1964). A related parasite, *Euryhalmis monorchis* has been reported in mink in Michigan (Ameel, 1938). The mink is the natural host for *Euparyphium beaveri* in Michigan, while *Euparyphium inerme* has been reported to infect river otters in the Pacific Northwest (Miller and Harkema, 1964; Hoberg et al., 1997).

4.2. Cestodes of American marten in Michigan

A single American marten from the NLP was shedding *Hymenolepis* species ova. *Hymenolepis nana* is a zoonotic cestode of rodents, carnivores and humans found worldwide, but other species of *Hymenolepis* infect galliformes, including potential American marten prey species. The parasite may use intermediate hosts or paratenic hosts, including dung beetles, stable flies and fleas (Drew, 2003; Joslin, 2003; Loomis, 2003; Sainsbury, 2003). Without knowing the species of *Hymenolepis* found from the American marten in the NLP, it is not known whether this may have been a pass-through finding or was truly an infection of the American marten; the zoonotic potential is not known.

4.2.1. Other reported cestodes of American marten in North America

Taenia species are cestodes (commonly known as tapeworms) that maintain a completely sylvatic life cycle. The definitive hosts for *Taenia mustelae* and *Taenia martis americana* are primarily mustelids. Adult parasites live in the small intestine and eggs are passed in the faeces of the host. Larvae hatch in the environment and are ingested by herbivores, the intermediate hosts. The ingested larvae form cysticercus in skeletal muscle and viscera, and the life cycle is completed when a carnivore consumes the infected intermediate host. While the definitive host typically shows no signs of disease, the intermediate host may suffer morbidity or mortality, including liver damage, as a result of infection (Jones and Pybus, 2001).

T. mustelae has a wide distribution across the Northern hemisphere and has been reported in North American mustelids,

including American marten, short-tailed weasel (*Mustela erminea*), mink (*Neovison vison*) and least weasel (*M. nivalis*). The cestode has been found in 2 sciurid definitive hosts, *Marmota broweri* and *M. caligata* (Jones and Pybus, 2001). *Taenia martis americana* infects mustelids, including American marten, fisher (*Martes pennanti*) and the ringtail (*Bassariscus astutus*, Family: Procyonidae), as definitive hosts. Rodents in North America reported with the larval stage of infection include *Lemmus sibiricus*, *Microtus xanthognathus*, *Mus musculus* and *Ondatra zibethicus* (Jones and Pybus, 2001). Two American marten from the southern Cascades were co-infected with both *T. mustelae* and *T. martis americana* (Hoberg et al., 1990).

Mesocestoides species are cestodes with a complicated life cycle. Adult parasites are found in the small intestine of definitive hosts, which include canids, felids and mustelids (Bowman et al., 2009). Larval or adult parasites can also infect birds, reptiles and other mammals (Wardle and McLeod, 1952). Eggs, or gravid proglottids, are suspected to be ingested by a first intermediate host, a coprophagic insect or a mite (Chowdhury and Aguirre, 2001; Bowman et al., 2009). The insect is consumed by a second intermediate host, which may include birds, mammals, reptiles and amphibians. Lastly, the definitive host becomes infected by ingesting the second intermediate host (Bowman et al., 2009). Definitive hosts may have clinical signs of infection including anorexia, low serum albumin and vomiting (Chowdhury and Aguirre, 2001). Humans can be incidental definitive hosts and become infected by consuming undercooked game (Chowdhury and Aguirre, 2001; Fuentes et al., 2003). *Mesocestoides lineatus* was identified in the small intestine of 33% (n = 14) of American marten from eastern Washington with significantly higher rates in juveniles than in adults. Current taxonomy, however, suggests that this species may have actually been *M. variabilis*, which occurs in North America, while *M. lineatus* is an Old World species (Fuentes et al., 2003). Coinfections with *M. lineatus* and *Capillaria putorii* occurred in 35% of parasitized American marten, and juveniles had statistically higher rates of coinfection than adults (Foreyt and Langerquist, 1993).

4.3. Nematodes of American marten in Michigan

Hookworms are zoonotic nematode parasites infecting carnivores (Taylor et al., 2007). Carnivores may become infected via ingestion of larvae or eggs, paratenic hosts, or via percutaneous or lactogenic transmission. Larvae migrate to the lungs, moult, and are coughed up and swallowed to lay eggs in the small intestine. Ingested larvae may bypass pulmonary migration or migrate out of the lungs into the muscle and remain in an infected female mammal until pregnancy occurs, at which point the larvae migrate to the mammary gland leading to lactogenic transmission (Taylor et al., 2007). Hookworm infection in dogs and foxes can result in bloody diarrhoea, anaemia, poor hair coat, poor growth in puppies and respiratory signs (Taylor et al., 2007).

We found that the odds of having anaemia (haematocrit <42% as described by Nieminen et al., 2007) were 8.75 higher for American marten infected with hookworms than in uninfected American marten in this study. While there was a significant association between presence of hookworm infection and presence of anaemia in this report, other clinical signs related to hookworm infection were not seen. Because other causes of anaemia were not investigated, the association does not prove causation and warrants further investigation. In addition, molecular techniques could be used to determine the species of parasite infecting American marten in Michigan. Because of the potential for anaemia to affect fitness of an individual American marten or the growth of kits infected via transmammary transmission, treatment of hookworm-infected American marten destined for relocation may be warranted.

Capillaria is a genus encompassing many species. *Aonchotheca putorii* (previously known as *Capillaria putorii*) infects the gastrointestinal tract of mustelids and other species, while other capillarids infect the respiratory tract, bladder or liver of their respective definitive hosts (Bowman et al., 2009). *A. putorii* can have a direct or indirect life cycle in which adult parasites shed eggs in the gastrointestinal tract of the mammalian host. The eggs are capable of infecting other susceptible hosts directly or using an earthworm as an intermediate host (Segovia et al., 2007; Taylor et al., 2007). *A. putorii* was found in 77.8% (n = 14) of American marten from the UP in the current report, which was significantly higher than the 29.0% (n = 9) prevalence in the NLP (P < 0.05). However, during the initial stage of this parasite survey, some *Capillaria* ova were not identified to species, and these are represented as “unidentified *Capillaria*” in Table 1. If the 42.0% (n = 13) prevalence of unidentified *Capillaria* found in the NLP were in fact *A. putorii*, then there would not be a significant difference in prevalence of *A. putorii* between the UP and NLP. The prevalence of *A. putorii* from the UP in this report is higher than the 47% (n = 66) previously reported from American marten carcasses from the UP (Veine-Smith et al., 2011). *A. putorii* has also been reported in the stomach of ferret, mink, short-tailed weasel, raccoon, fisher and striped skunk (*Mephitis mephitis*) and in the small intestine of bobcats, bears, raccoons, swine, hedgehogs and the domestic cat (Foreyt and Langerquist, 1993; Bowman et al., 2009). In northeastern Washington, *C. putorii* was found in 86% (n = 13) of American marten carcasses, mostly in the stomach and less frequently in the large or small intestine of American marten. Using faecal flotation alone, *Capillaria* species ova were found in 64% (n = 21) of samples examined from the same population (Foreyt and Langerquist, 1993). Additional future comparisons between carcass and faecal examinations to determine prevalence of *Capillaria* and other parasites is warranted.

Eucoleus aerophilus, previously known as *Capillaria aerophila*, infects the respiratory tract of hosts including American marten and other carnivores, such as fisher, red fox (*Vulpes vulpes*), raccoon, coyote (*Canis latrans*), striped skunk and badger (*Taxidea taxus*) (Bowman et al., 2009). *E. aerophilus* rarely infects humans (Lalošević et al., 2013). The life cycle of *E. aerophilus* may be direct or indirect with earthworms serving as intermediate hosts (Bowman et al., 2009). In farmed foxes, infection with *E. aerophilus* can lead to respiratory disease and clinical signs include coughing, wheezing, failure to thrive, pneumonia and even death. Cats and dogs have also been infected but typically do not suffer the same degree of clinical signs as foxes since their infections are not as intense (Bowman et al., 2009). *E. aerophilus* was identified in 4% of the respiratory tracts of American marten from Ontario, but it is unknown whether infection resulted in disease or increased risk of being trapped (Seville and Addison, 1995). *Eucoleus boehmi* has been reported to infect the respiratory tract of foxes and dogs (Bowman et al., 2009). Its ova can be differentiated from the similar *E. aerophilus* by its pitted surface (Bowman et al., 2009). The relative contribution of earthworms to the American marten diet in Michigan is not known but may be significant given the high overall prevalence of capillarid parasites seen there.

Physaloptera species are found in the stomach of infected carnivores, including mink, striped skunk, raccoons, dogs, and cats, and eggs are shed intermittently (Chowdhury and Aguirre, 2001; Veine-Smith et al., 2011). Crickets, beetles or other invertebrates act as intermediate hosts, while rodents and reptiles may be paratenic hosts (Chowdhury and Aguirre, 2001). While most infections do not cause disease in the host, severe ulcerative gastritis has been reported in the bandicoot, a marsupial (*Perameles* species) (Holz, 2003).

Crenosoma species is a lungworm found within the respiratory

tract of carnivores and insectivores (Craig and Anderson, 1972). Adult parasites lay eggs in the lungs; larvae are coughed up, swallowed, and passed in host faeces. Larvae penetrate a gastropod, the intermediate host. The life cycle is completed when a carnivore consumes a snail or slug. Heavy infection with *Crenosoma* species can cause clinical signs such as coughing, sneezing, nasal discharge and difficulty breathing (Chowdhury and Aguirre, 2001). The red fox is the typical host for *Crenosoma vulpis* and is sympatric with American marten in both the UP and NLP. Given the global distribution of *C. vulpis*, it is likely that this parasite exists in the NLP red fox population although the parasite was not identified in American marten from the NLP in this study. *Crenosoma petrowi* has been reported from free-ranging Russian sable, a captive fisher in the United States and a badger from Canada (Craig and Anderson, 1972). A single American marten from Ontario was found to be infected with *Crenosoma petrowi* (<1% prevalence), but the reported prevalence in fisher from the same region was 15% (Seville and Addison, 1995). Olsen (1952) examined 62 carcasses of *Martes caurina* from Colorado and found 18 (29%) to be infected with *Crenosoma*, which he designated *Crenosoma coloradoensis*. *Crenosoma* species was found in the lungs of 2% (n = 3) of American marten from the UP of Michigan (Veine-Smith et al., 2011).

Strongyloides martis and *S. lutrae* have been reported in river otters (Hoberg et al., 1997). Parasites of this genus are generally species or host-specific and undergo both a direct life cycle and a free-living stage. Infective larvae or eggs in the soil are consumed by the host; larvae of some species can also enter the host through the skin (Morris and Shima, 2003). Pathogenicity of *Strongyloides* species in mustelids is not known, but disease could result from migration of the parasite through the lung (Kimber and Kollias, 2000).

Diocotylus renale, commonly known as the giant kidney worm, is one of the largest roundworms infecting wild and domestic species worldwide including wolves, bears, foxes and mink, as well as domestic dogs, cattle, horses and pigs. Humans have also been reported with *D. renale* (Chowdhury and Aguirre, 2001). The adult worm is typically found in the right kidney because the infective larvae exits the intestinal tract on the right side near the stomach. The parasite can live up to 3 years in the kidney. When the parasite dies, the kidney is essentially destroyed, and the host becomes reliant on the remaining left kidney. Occasionally, both kidneys are infected or the parasite is found elsewhere in the abdomen. An adult female worm lays eggs within the kidney, and the eggs are shed in the urine of the host mammal. It takes about 6 months for the egg to become infective, at which point it may be swallowed by the intermediate host, *Lumbriculus variegatus*, an aquatic annelid commonly known as blackworm. The egg develops within the intermediate host into an infective larvae. If the annelid is eaten by an American marten, or other mammalian host, the life cycle is completed when the larvae finds its way to a kidney. Fish, frogs and crayfish may act as paratenic hosts by consuming the infected annelid (Cheng, 1986; Chowdhury and Aguirre, 2001). A single American marten from the UP was reported with nephritis due to suspected prior infection with *D. renale* (Spriggs, unpublished data). In an examination of 405 American marten from Ontario, *D. renale* was found in only 2% of American marten and only from districts with previous reports of infected mink. In 4 of the 5 infected American marten, there was evidence only of past infection such as the entire right kidney missing or merely a fibrous capsule remaining, while in 1 American marten the actual parasite was identified (Seville and Addison, 1995).

Filaroides martis is a helminth parasite found in the trachea, bronchi and lungs and has been reported to infect mustelids, including mink and American marten, as well as canids (Chowdhury and Aguirre, 2001). Aquatic and terrestrial snails are

the intermediate hosts. The larvae moult in the stomach mucosa of the definitive host and migrates to the thoracic cavity over the next month. Larvae increase in size over 100-fold during this timeframe (Ko and Anderson, 1972). Infection with *F. martis* has been reported to cause pneumonia in other species, but its effect on American marten is not known (Chowdhury and Aguirre, 2001). Of 405 American marten examined from Ontario, 8% had lung or aortic nodules associated with the parasite. Because yearlings had a significantly higher prevalence of infection than other ages, the authors suggest that infection could have an effect on survival of yearlings or that American marten are able to recover from infection at older age groups (Seville and Addison, 1995). *F. martis* was found in the lungs of 4% (n = 5) of American marten carcasses from the UP of Michigan (Veine-Smith et al., 2011). Histopathology revealed lesions consistent with verminous pneumonia in 60% (n = 9) of American marten carcasses from Michigan (Spriggs, unpublished data). While these were incidental findings and the causative parasite was not identified, it is possible that pneumonia may have resulted in mild respiratory compromise in affected American marten.

4.3.1. Other reported nematodes of American marten in North America

Pearsonema plica, previously known as *Capillaria plica*, was identified in the urinary bladder of 6% of American marten from Ontario (Seville and Addison, 1995). *P. plica* has been reported in the urinary tract of the domestic cat, dog, raccoon, red fox, coyote, wolf, striped skunk and fisher, as well as American marten (Butterworth and Beverley-Burton, 1980). The definitive host begins to shed eggs of *C. plica* in the urine about 8 weeks after consuming an earthworm, the paratenic host (Bowman et al., 2009). Infection usually does not cause disease for the host, but there is a suggestion that *P. plica* resulted in poor growth in fox kits (Bowman et al., 2009).

Baylisascaris devosi is a nematode reported in both American marten and fisher (Kazacos, 2001). Eggs shed from the definitive host become infective after 11–14 days, at which point small mammals such as rodents and squirrels become infected by ingesting the eggs while foraging. The larvae migrate throughout the tissues of these paratenic hosts and typically localize to the muscle of the forelimbs and thorax. Neural larval migrans, a neurologic disease, is rare or non-existent with *B. devosi* (Kazacos, 2001). In contrast, the larvae of the related *Baylisascaris procyonis*, or raccoon roundworm, frequently migrate to the central nervous system of non-adapted hosts and cause neural larval migrans. Humans are susceptible to neural larval migrans caused by *Baylisascaris* species, but *B. devosi* is less likely to cause disease in humans than *B. procyonis* (Kazacos, 2001). Adult *B. devosi* inhabits the small intestine; the definitive host does not typically show signs, but intestinal blockage is possible with a severe infection (Chowdhury and Aguirre, 2001). In 42 *M. caurina* carcasses examined from Idaho, one was infected with *B. devosi* (Erickson, 1946; Poole et al., 1983).

Soboliphyme baturini, commonly known as stomach worm, is a nematode parasite distributed from central Siberia across Beringia to the Pacific Northwest of the United States (Koehler et al., 2009). The primary definitive hosts are sable (*Martes zibellina*) and American marten. Other mustelids reported with the parasite include ermine (*M. erminea*), mink and ferrets (*Mustelo putorius furo*) (Levine, 1968; Swartz, 1968; Koehler et al., 2009). *S. baturini* has been reported in other carnivores, including the fox and domestic cat (Levine, 1968). Mature female worms are found in the stomach or small intestine of the mustelid host, and eggs are passed in the faeces of the host. Earthworms are the intermediate host, while shrews become paratenic hosts when they ingest the earthworm. American marten may be infected by consuming either

infected shrews or earthworms (Koehler et al., 2009). Koehler et al. (2009) used genetic molecular data of *S. baturini* to shed light on the expansion of the ancestral American marten across Beringia into North America, its speciation during isolation in glacial refugia, and re-colonization in Alaska and reinfection with *S. baturini*. Clinical manifestation of infection in sable includes anaemia and gastric ulceration (Thomas et al., 2008). There was a 55% (n = 155) prevalence of infection with *S. baturini* in American marten from Prince of Wales Island in Alaska (Table 3). There was no correlation between intensity of *S. baturini* infection and fat deposits which measured to assess nutritional condition (Thomas et al., 2008). American marten carcasses were collected over an 8-year time period from 3 locations in Alaska and stomachs were examined for *S. baturini* (Table 3). None of the American marten in that study had any sign of negative health impact from the parasite infection (Zarnke et al., 2004). A study conducted in Idaho examined 42 *M. caurina* carcasses and found one with *S. baturini* (Erickson, 1946).

Trichinella spiralis has a broad host range, infecting over 100 species of mammals. It is found worldwide except in Antarctica and Australasia (Chowdhury and Aguirre, 2001). Adult *T. spiralis* are found in the small intestine, and the females give birth to larvae which migrate through the body to become encysted in skeletal muscle. An American marten may become infected by consuming a rodent or other mammal with the encysted parasite. Once consumed, the larvae are freed and migrate to the small intestine of the host to complete their life cycle. Humans can acquire trichinellosis by eating undercooked meat (Taylor et al., 2007). The first report of *T. spiralis* in American marten in North America was from Manitoba, Canada, where a single yearling was found infected of 139 American marten examined (Poole et al., 1983). Prevalence of *Trichinella* in American marten and other species may vary from year to year (Dick et al., 1986). American marten from the northern and southern Cascades of Washington were found to have *Trichinella* encysted in the diaphragm, with a prevalence of 50% and 31%, respectively (Hoberg et al., 1990). Another study examined tongue muscle from 42 American marten from northeastern Washington and found only 5% prevalence, which the authors attribute to differences in technique and tissues examined (Foreyt and Langerquist, 1993). A study conducted in Ontario found a 3.4% prevalence (n = 68) of infection with *T. spiralis* in American marten (Dick et al., 1986). In that study, 4773 carnivores of 18 species were examined. Only American marten, fishers and mink were found with the parasite. The prevalence in fishers in the study was slightly higher at 4.5% (n = 83), and a single mink (of 12 tested) was positive. The authors suggested that American marten and fishers are key in the sylvatic transmission of *Trichinella* in this part of Canada (Dick et al., 1986). Other reports of *T. spiralis* in American marten are found in Table 3. In contrast to the findings of Dick et al. (1986) in which only mustelids were found with *T. spiralis*, this study found a high prevalence in coyote (61%, n = 22) and confirmed infection in a variety of other mammals including carnivores and rodents (Schmitt et al., 1976).

Dracunculus insignis is a parasite known to infect raccoons, dogs, mink, fishers and skunks in the United States east of the Rocky Mountains and in Ontario, Canada (Crichton and Beverley-Burton, 1973; Cheng, 1986). Experimental infections in ferrets are used as a model for human dracunculiasis (Eberhard et al., 1988). *Dracunculus lutrae* infects otters in North America (Crichton and Beverley-Burton, 1973). Both species have a similar life cycle to the related the Old World parasite *Didelphis medinensis*, commonly called the guinea worm, a zoonotic parasite. The adult worm is found in abdominal tissues of the host. Female *D. lutrae* migrate to the tissues under the skin of the animal to give birth to live larvae while secreting a substance that causes a blister to form. When the blister ruptures and is exposed to water, the infective larvae is

released into the water. The free-living larvae can survive for several days until they are consumed by an aquatic copepod, the intermediate host. *D. insignis* develops through several more stages within the copepod over the next 3 weeks. When a definitive host ingests the copepod while drinking water, the larvae are freed in the stomach or small intestine and migrate to abdominal organs and tissues where they continue to develop into adult worms over the next 8–12 months (Cheng, 1986). *D. insignis* was found in only a single American marten of 405 examined in Ontario despite the parasite being commonly found in raccoons of the same region. The authors suggested a higher true prevalence, but detection was low due to the pelt being removed for commercial reasons in most of the examined specimens (Seville and Addison, 1995). This parasite is of economic concern as it can affect pelt quality in fur-bearer species.

4.4. Protozoa of American marten in Michigan

A single American marten from the NLP was found to be shedding *Sarcocystis* species sporocysts. *Sarcocystis* species is a protozoal parasite that has infrequently been reported in carnivores such as domestic cats, dogs, raccoons, cougars, bobcats, mink, striped skunks, sea otters, fishers and Pacific harbor seals (Foreyt and Langerquist, 1993; Gerhold et al., 2005; Larkin et al., 2011). *Sarcocystis* species require an intermediate host. Gerhold et al. (2005) reported a case of meningoencephalitis in a fisher caused by *Sarcocystis neurona* in Maryland, USA. The Virginia opossum (*Didelphis virginiana*) is the definitive host for the parasite in North America, but the natural intermediate host for *S. neurona* has not been discovered (Gerhold et al., 2005). *Sarcocystis* species was found in the tongues of 10% ($n = 4$) of American marten examined from northeastern Washington (Foreyt and Langerquist, 1993).

Antibodies to *Toxoplasma gondii* were detected in 10.8% ($n = 15$) of American marten in Ontario. American marten in Michigan were found to have a 58% seroprevalence ($n = 47$), and there was no significant difference between the UP and NLP (Spriggs, unpublished data). *T. gondii* has been reported to cause mortality in the related black-footed ferret (*Mustela nigripes*), captive-raised American mink (*Neovison vison*) and southern sea otters (*Enhydra lutris*) (Dubey et al., 2003; Burns et al., 2003; Jones et al., 2006).

Coccidian parasites have been only rarely reported in American marten, but this is likely because few studies have used faecal flotation and/or histopathology to detect parasites. All coccidian parasites are obligate, intracellular parasites and undergo stages of asexual and sexual reproduction in the life cycle, which may be direct or indirect. Coccidian oocysts from 5 American marten in this report were sporulated to allow identification to the genus level. *Eimeria* species were identified in 2 of the 5 sporulated samples from American marten in the current study, and *Cystoisospora* species in the remaining 3 samples. It is unknown whether coccidian oocysts seen in faecal samples were pass-through from prey species. Taxonomy of coccidian parasites has been controversial; some have considered *Cystoisospora* species to be synonymous with *Isospora* species, while others believe it to be a distinct genus based on molecular and morphological characteristics (Yi-Fan et al., 2012). *Cystoisospora* species were identified in American marten from Michigan in this report based on morphological features of the sporulated oocysts. Yi-Fan et al. (2012) reported *Cystoisospora* species in steppe polecats in China and suggested that previously reported *Isospora* species in various other mustelids, including that of sable, should be reassigned to *Cystoisospora* species. Faeces from 33 American marten from eastern Washington were examined, and 6% were found with coccidian oocysts (Foreyt and Langerquist, 1993). *Cryptosporidium* and *Giardia* species cysts were detected in the faeces of American marten from the UP of Michigan, both

having a prevalence of 4% ($n = 5$) (Veine-Smith et al., 2011).

5. Conclusion

North American marten are infected with a wide variety of endoparasites, but information regarding the pathologic effects of parasitism remains limited. American marten infected with hookworms in the current study were found to be at risk for anaemia, and this association warrants further investigation. Some parasites are infrequently reported in only a single location, while those reported in multiple locations typically have a varying prevalence. Multiple ecological factors, including habitat, prey availability, sympatric carnivore community, and host adaptation, are likely involved in the variation of prevalence at different geographic locations. A number of parasites known to infect American marten have zoonotic potential. As American marten are frequently trapped as a furbearer species, this information could guide prevention of disease transmission to trappers and researchers working with the species.

Parasite infections could cause illness under conditions of stress and presumed immunosuppression associated with reintroduction programs (Kimber and Kollias, 2000). The majority of previous studies examined American marten carcasses for parasitism, which allows characterization and speciation of the adult parasite and information about intensity of infection. However, a non-invasive method of detecting endoparasitism is desired for reintroduction programs and our results provide baseline information about parasites detected by faecal examination from American marten in Michigan. Multiple faecal exams from individuals destined for relocation, including sedimentation and molecular techniques when available, are warranted to identify novel parasites and parasites with pathologic or zoonotic potential and to make appropriate treatment or management decisions.

Acknowledgements

We gratefully acknowledge field personnel, particularly B. Sanders, C. Schumacher, T. Hillman, and M. Nichols. We thank J. Witt, P. Keenlance, D. Miller, and R. Wilkes for their comments on this manuscript and R. Thornton for the American marten photograph. We gratefully acknowledge the Evansville Zoological Society, Ulysses S. Seal Conservation Grant through the Minnesota Zoo, and the PPG Sustainability & Conservation Fund through the Pittsburgh Zoo & PPG Aquarium for financial support. We acknowledge the Little River Band of Ottawa Indians Natural Resource Department, Sault Tribe of Chippewa Indians Natural Resource Department, Keweenaw Bay Indian Community Natural Resource Department, and the Detroit Zoo for their contributions to the fieldwork.

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