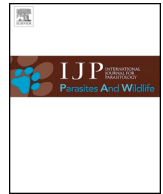




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A volunteer-populated online database provides evidence for a geographic pattern in symptoms of black spot infections

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ABSTRACT

Infections of parasitic digenean trematode metacercariae may lead to a visually observable syndrome in fish commonly called black spot disease. While black spot has been noted from various locations throughout North America, patterns in prevalence across the continent remain unknown. Funding to investigate continental-wide prevalence of low-mortality parasitic infections represents a barrier to such studies. I utilize iNaturalist.org's photograph database to examine fish for signs of black spot infections across North America. Fish targeted include blacknose dace, creek chub, chubs (*Nocomis* spp.), and stonerollers (*Camptostoma* spp.). Photos were visually examined for symptomatic black spots indicative of infection by trematode species linked to black spot disease. Regardless of fish species group, symptoms of black spot pathogens were highly prevalent (27.1% of 314 fish) in watersheds of southern Ontario Canada, whereas mean prevalence was comparatively low elsewhere (7.8%). In one instance, a user uploaded a higher number of photos, with a higher percentage exhibiting signs of infection than other users in the watershed. However, it is difficult to tease apart if that user fished in waterbodies with high infection rates, uploaded more photos of symptomatic fishes, or some other explanation for the differences in user-reported fish with symptoms. Beyond this exception, geographic patterns in the frequency of black spot symptoms do not appear to be related to solely the users, suggesting the observed pattern is biological or ecological. While causative explanations remain conjectures, the data reported herein provides evidence that across four groups of fish, signs of black spot infections are more common in southern Ontario than other areas studied in North America. This work also represents an initial and unexpected utility of volunteer-population databases such as iNaturalist. Further data contributions could lead to better understanding of the causative agents to variation in black spot pathogens' occurrences.

1. Introduction

Black spot disease, or black grub disease, is a visually observable infection caused by schistosome-like digenean parasites of multiple trematode genera among the families Diplostomatidae and Heterophyidae, most commonly reported as *Apophallus* spp., *Crassiphiala* spp. or *Uvulifer* spp. (Hoffman, 1999) among others, or more generally as neascus-type trematodes (Cairns et al., 2005). The name black spot disease refers to visible melanocytes that form around the location where metacercariae encyst in fish species' integument, musculature, or fins (Duru et al., 1981). Due to this etiology, the symptomatic black spots are well defined and thus rather distinct from melanocytes that may occur around trauma (i.e., cuts, scrapes, and punctures) or natural coloration and can be seen with the naked eye. Life cycles of these parasites are rather complex, utilizing multiple host species including snails (often Planorbidae), a secondary or intermediate host fish species, and a definitive host species of piscivorous

bird, for example belted kingfisher (*Megaceryle alcyon*) is often cited (Lane and Morris, 2010). Host fish species can differ by trematode species (Hoffman and Putz, 1965; Locke et al., 2010), but black spot disease is commonly reported in members of Cypriniformes, Esociformes, Perciformes, and Salmoniformes (Cairns et al., 2005; McAllister et al., 2013; Wisenden et al., 2012).

Although black spot disease has long been known in scientific literature (Hunter and Hamilton, 1941), most studies on its prevalence focus on fine-scale geographic areas (Berra and Au, 1978; Blouin et al., 1984; McAllister et al., 2013; Quist et al., 2007). These studies indicate a northern range of southern Canada (Steedman, 1991), south into Arkansas (McAllister et al., 2013) and Texas (Tobler et al., 2006; Tobler and Schlupp, 2007), west into Oregon (Cairns et al., 2005), and along the eastern coastline of the United States (Lemly and Esch, 1984). Although the parasites are thought to occur throughout much of North America, a continental-wide survey of black spot disease has yet to be reported.

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Lack of funding for parasitology research has been identified as one of largest barriers for studies, especially for parasites where economic impacts are low or unknown (Scholz and Choudhury, 2014). Mortality caused by black spot disease appears species-specific, with reports on increased over-winter mortality for bluegill (*Lepomis macrochirus*; Lemly and Esch, 1984; Pracheil and Muzzall, 2010), reduced survival of young northern pike (*Esox lucius*; Harrison and Hadley, 1982) and shortnose suckers (*Chasmistes brevirostris*; Markle et al., 2014). However, others have indicated no effects on mortality of yellow perch (*Perca flavescens*; Vaughan and Coble, 1975) or fathead minnows (*Pimephales promelas*; Wisenden et al., 2012), nor on thermal tolerances of cyprinids (Hockett and Mundahl, 1989). Without clear and direct mortality consequences, negative effects on fish hatcheries, nor largescale outbreak events to bring attention to the disease, black spot disease has received little continent-wide attention.

Data voluntarily provided by community members, often referred to as citizen science, offers a means of collecting data at frequent intervals and across wide geographic areas with a low cost to agencies. As of July 2019, the iNaturalist database hosts over 23.6 million publicly accessible observations of organisms. Observations on iNaturalist include georeferenced photographs of organisms and community-sourced species identifications. Similar community-member provided observational data have offered updated species distribution ranges (Suzuki-Ohno et al., 2017), knowledge on species' migrations (Hurlbert and Liang, 2012; Soroye et al., 2018; Walker and Taylor, 2017), evidence of abundance and diversity changes (Clark, 2017; Gutiérrez-Tapia et al., 2018), and even information on the volunteer's themselves (Kolstoe and Cameron, 2017; Papenfuss et al., 2015). As such, the scientific community is beginning to recognize the power and utility of such online databases as tools for research that would have previously lacked funding. Herein I report on the use of the iNaturalist database (iNaturalist.org, 2019) to investigate geographic patterns of fish exhibiting signs of black spot related trematode infections.

2. Methods

Photos of blacknose dace (*Rhinichthys atratulus* & *R. obtusus*), chubs (*Nocomis* spp.), creek chub (*Semotilus atromaculatus*), and stonerollers (*Campostoma* spp.) were visually inspected on iNaturalist.org for evidence of black spot metacercaria, specifically on fins, torso, and the ventral side of gill structures. Fish species were chosen based on multiple factors including the number of observations for each species on iNaturalist, a wide geographic distribution of those observations, species of small-bodied fishes allowing easier classification of black spot parasite presence (vs. for example large specimen of bluegill *Lepomis macrochirus* or yellow perch *Perca flavescens*), and species known to commonly be infected by black spot pathogens (Evans and Mackiewicz, 1958). Members of the chub and stoneroller groups are often difficult to identify to species which, when combined with the low number of observations for individual species within each group compared to blacknose dace or creek chub, made it more convenient to lump these species at the genus level (*Campostoma* and *Nocomis*). Other species, such as *Esox* spp., *Lepomis* spp., *Perca* spp., and Salmonine spp. have iNaturalist observations spanning a comparatively large range in sizes and were thus excluded as the black spots become more inconspicuous with larger sizes and since observations of larger individuals were often too far from the camera for accurate examination for infections compared to smaller conspecifics.

Photos of each fish were examined for visually evident well-defined black spots on fish that were inconsistent with known natural coloration patterns and variations (Fig. 1) and classified as either “infected” (presence of signs of black spot disease) or a “noninfected” (absence of signs of black spot disease). The term “infected” is used herein to represent cases with externally visible signs of metacercaria and does not denote other internal pathologies nor were cysts dissected for confirmation. Black spots were distinguished from dark debris primarily by

shape and size, with the parasites being nearly perfectly circular compared to leaf or woody debris on fishes, which were also usually evident on hands or containers fish were in allowing comparison. When in doubt between debris, natural coloration, or evidence of black spot, error was towards non-infected. Photos which contained the fish but could not be confidently observed for evidence of black grub (i.e., not close enough to lens, out of focus, free swimming, torso covered by hand, or too coated in debris) were excluded from the dataset (61 observations of 1444).

Records were restricted to observations posted since Jan 1st 2015, had at least one photo, and had GPS locations available whether obscured or open. Photos taken prior to 2015 were comparatively sparse and geographically dispersed and thus this date served as a cutoff point after which data was collected and used. Observations marked as private on iNaturalist did not offer GPS locations and thus were not obtained nor enumerated. When a user chooses to obscure GPS coordinates in iNaturalist, a randomized coordinate pair within a 0.2 by 0.2° area that contains the true coordinates is stored. The coordinates of the randomized point were assumed to still lie within the watershed of interest and thus were retained, this occurred for 64 of the 1383 observations classified as either “infected” or not.

Watershed polygons created by the Commission for Environmental Cooperation were obtained from USGS ScienceBase and consisted of coarse watersheds for larger rivers (HUC 6 for U.S. and Sub-Basins for Canada; CEC, 2011). Watershed polygons were overlain on a map of North America obtained from Stamen Maps in rStudio using *ggmap* (Kahle and Wickham, 2013). Only watersheds in which at least one observation occurred were retained for each of the four fish groups being mapped. The watershed's polygon fill color was used to denote the proportion of fish classified as “infected” within each watershed. Fill colors range from black (0% infected) to yellow ($\geq 50\%$ infected). To account for differences in the number of observations, watersheds were grouped by the number of observations for each species: those with only 1 or 2 observations were made transparent; those with 3–5 observations were set to 20% opaque; those with 6–10 observations were set to 50% opaque; those with 11–15 were set to 75% opaque; and polygons with > 15 observations were set to be fully opaque. As such, stronger colors mean more observations whereas brighter colors mean a higher percentage are classified as infected.

To assess the possibility that certain users only took photos of fish displaying black spot parasites, data were summarized for the 15 users whom posted at least 10 total observations. I also summarized the observations exhibiting signs of infection by year and watershed for users that reported more than 3 fish with visual signs of infection. Further, data from three watersheds in southern Ontario were summarized by year. These summaries were done to tease apart the interplay of users, year, watershed, as well as the number users in a watershed each year.

To assess which variables describe data, and thus the probability of exhibiting signs of infection, conditional inference trees were trained and evaluated using the *train* function in R programming package *caret* (Kuhn, 2015, 2008). Predictors included were Year, Common Name of the species or group (i.e., chubs and stonerollers), User name, and Watershed of capture. Models were evaluated with *train* using Monte Carlo Cross Validation with a randomly selected 75% of the data used as training set over a selection of 15 different values for the decision criterion tests each of 20 iterations. The model with the highest classification accuracy was retained and the decision tree plotted. Variables included in the final model were deemed as most important in explaining the data.

3. Results

I inspected photos of 306 blacknose dace, 248 chubs (*Nocomis* spp.), 553 creek chub, and 276 stonerollers (*Campostoma* spp.) on iNaturalist for evidence of black spot parasite infections (Fig. 1). Of which, 12.1% (37) of blacknose dace, 19.8% (49) of chubs, 9.5% (53) of creek chub,



Fig. 1. Example photos of a blacknose dace, a stoneroller (*Campostoma* sp.), a creek chub, and a chub (*Nocomis* sp.) exhibiting evidence of a black grub infection.

Table 1

Numbers of species caught and posted to iNaturalist in each watershed and, in parentheses, the percentage of those fish that had visible signs of black spot infections. Included is the total percentage of fish in each watershed infected (Total Infect.) and the number of users contributing data. Data limited to watersheds with ≥ 10 observations.

Watershed	Blacknose Dace	Campostoma spp.	Creek Chub	Nocomis spp.	Total Infect.	# Users
Lake Ontario and Niagara Peninsula	60(15)	9(11.1)	88(19.3)	33(30.3)	19.5	54
Southern Lake Erie	31(6.5)	35(0)	47(8.5)	1(0)	5.3	15
Potomac	46(6.5)	7(0)	20(10)	4(25)	7.8	41
Northern Lake Erie	10(50)	19(68.4)	23(26.1)	12(66.7)	50	17
Eastern Georgian Bay	13(30.8)	2(50)	28(10.7)	17(47.1)	26.7	12
Coosa-Tallapoosa	1(100)	7(0)	24(8.3)	6(0)	7.9	11
Middle Tennessee-Elk Little	5(40)	5(0)	18(0)	1(0)	6.9	7
Little	-	26(0)	-	-	0	1
Middle Colorado-Llano	-	26(0)	-	-	0	15
French Broad-Holston	9(22.2)	7(0)	1(0)	7(28.6)	16.7	13
Central St. Lawrence	6(0)	-	17(5.9)	-	4.3	7
Neuse	-	-	3(0)	18(0)	0	7
Upper White	-	3(0)	2(0)	15(0)	0	9
Middle Tennessee-Hiwassee	2(0)	5(0)	8(12.5)	5(0)	5	8
Upper Pee Dee	-	-	8(0)	11(9.1)	5.3	7
Apalachicola	-	5(0)	2(0)	11(54.5)	33.3	8
Santee	-	1(0)	6(0)	11(9.1)	5.6	9
Lower Tennessee	-	2(0)	12(8.3)	3(33.3)	11.8	2
Southeastern Lake Michigan	3(0)	-	12(8.3)	2(0)	5.9	4
Upper Hudson	10(0)	-	6(0)	-	0	9
Lower Missouri-Blackwater	-	5(0)	11(0)	-	0	8
Allegheny	2(0)	-	4(0)	10(10)	6.3	10
Eastern Lake Huron	2(0)	2(0)	9(0)	2(50)	6.7	10
Kanawha	3(0)	1(0)	4(0)	6(16.7)	7.1	10
Upper Chesapeake	7(0)	1(0)	3(0)	3(33.3)	7.1	11
James	6(0)	1(0)	1(0)	6(0)	0	9
Lower Delaware	7(0)	-	7(0)	-	0	8
Neosho	-	5(20)	3(0)	4(25)	16.7	6
Patoka-White	-	1(0)	11(0)	-	0	8
Richelieu	6(16.7)	-	6(0)	-	8.3	5
Lower Hudson	6(0)	-	5(0)	-	0	7
Upper Ohio-Beaver	4(25)	2(0)	3(0)	1(100)	20	9
Upper Tennessee	1(0)	3(0)	2(0)	4(0)	0	4
Scioto	2(0)	1(0)	7(14.3)	-	10	7
Upper Illinois	2(50)	5(20)	1(0)	2(0)	20	8

and 7.2% (20) of stonerollers were classified as “infected”. Of those showing signs of infection, 9 blacknose dace, 10 chubs, 17 creek chub, and 1 stoneroller were from the Lake Ontario-Niagara Peninsula watershed that surrounds Toronto, Canada (Table 1). A further 32 fish with signs of metacercariae infections were from the Northern Lake Erie watershed and another 16 from the Eastern Georgian Bay watershed, leading to the southern portion of the Province of Ontario Canada containing 53.5% of the 159 fish classified as “infected” in the dataset (Table 1). The average percentage of fish exhibiting signs of black spot infections of the three southern Ontario watersheds was 32.0% (the summed total was 27%; 85 fish out of 314), whereas the average across other watersheds, with at least 10 fish observations, was 6.8% (Table 1), and 7.8% across the whole dataset (inclusive of southern

Ontario). In contrast to southern Ontario, the Southern Lake Erie watershed represented many observations but a relatively low percentage of fish (5.3% of all 114 fish) exhibited neascus-type cysts. Geographic patterns of prevalence were generally similar across all species, with the southern portion of Ontario having higher proportions of fish with black spots than elsewhere where fish were encountered (Fig. 2). With one notable exception of 54.5% of the 11 chubs found in the Apalachicola watershed, which includes the Chattahoochee River that drains Atlanta, GA to the south.

Data were provided by 471 different user profiles, some of which were clearly avid anglers, or netters, of small-bodied fishes as the top 4 users caught over 18.3% of the 1383 fish included in the dataset (Table 2). Photos of fish exhibiting signs black spot metacercariae were

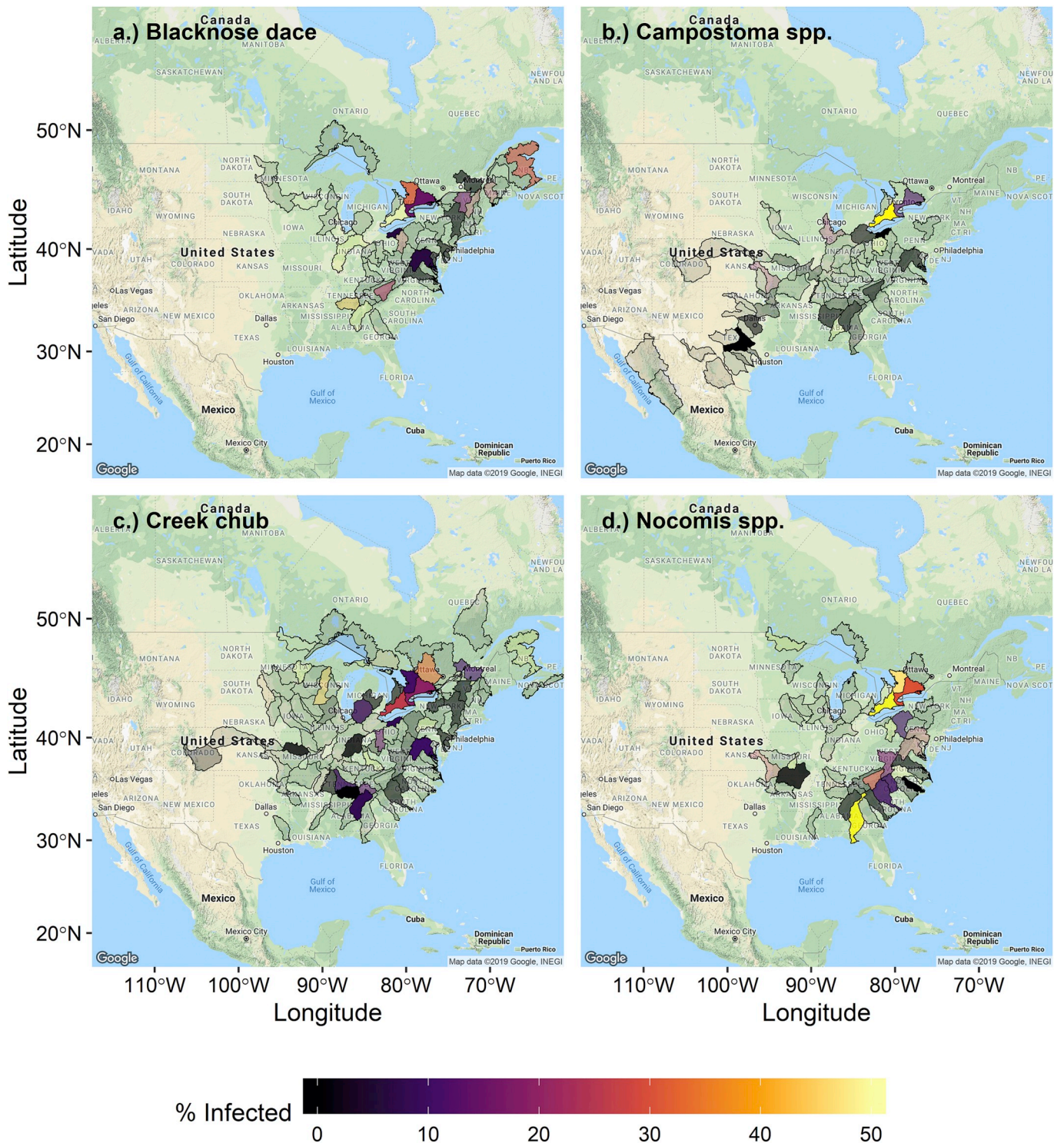


Fig. 2. Watersheds shaded by the percentage of observations with signs of black spot infection for four groups of fishes, each of which show higher prevalence in the watersheds near Toronto and the northern shoreline of Lake Erie. Watersheds are transparent if only 1 or 2 fish were observed in them, otherwise they increase in opacity as sample number increases, those with >15 observations are solid. Refer to Table 1 for numbers.

not isolated to any particular user and appear relatively evenly distributed across users (Tables 2 and 3). When stratified by user, year, and watershed, photos with evidence of infection appear to be from the same region (southern Ontario, CA) regardless of the user or the year fish were caught in (Table 3). Within this region, there was evidence that each year had increasing frequency of infections across each species group (Table 4).

The resulting decision tree included 3 splits and had an accuracy of

88.5%. Data from the Northern Lake Erie watershed, a specific user, or the Lake Ontario and Niagara Peninsula watershed were deemed more likely to have symptoms of black spot infections (Fig. 3). Of data from user *bkorol*, many (34 of 37) were from East Georgian Bay of which 12 exhibited signs of black spot infection, accounting for all but four fish exhibiting symptoms within this watershed. Eleven other users posted a total of 26 observations from East Georgian Bay, of which four (15.4%) exhibited signs of infections. As such, in this case, the user's ID was a

Table 2

Numbers of fish caught by iNaturalist users and, in parentheses, the percentage of those fish that exhibited signs of black spot infection. Included is the total percentage of fish infected by user (Total Infect.). Results limited to the 15 users with the most observations.

User	Blacknose dace	<i>Camptostoma</i> spp.	Creek chub	<i>Nocomis</i> spp.	Total Infect.
quiggifur	1 (0)	6 (0)	43 (2.3)	36 (16.7)	8.1
tortuga_rapido	25 (8)	28 (0)	31 (6.5)	–	4.8
fishesoftexas	–	43 (0)	–	–	0
oridgen10	15 (40)	3 (0)	15 (26.7)	7 (28.6)	30
bkorol	8 (37.5)	–	14 (14.3)	15 (53.3)	35.1
pmk00001	7 (57.1)	6 (0)	11 (18.2)	11 (36.4)	28.6
nv_multispeciesfishing	2 (0)	–	5 (0)	20 (5)	3.7
birds_bugs_botany	–	16 (75)	7 (28.6)	–	60.9
daniel_folds	2 (50)	10 (0)	8 (12.5)	3 (33.3)	13
rileywalsh	7 (0)	4 (0)	4 (0)	7 (42.9)	13.6
scottgibson	4 (25)	1 (100)	11 (36.4)	5 (40)	38.1
reuve nm	9 (0)	–	7 (28.6)	4 (50)	20
bradleyfishes	3 (0)	1 (0)	8 (12.5)	5 (60)	23.5
ncangling	–	4 (0)	3 (0)	9 (0)	0
flabellare223	3 (0)	2 (0)	4 (0)	6 (0)	0

Table 3

Counts of fish exhibiting black spot parasites summarized by user and the watershed the fish was captured in. Fish species included blacknose dace, *Camptostoma* spp., creek chub, *Nocomis* spp. Data displayed are limited to those users who posted > 3 observations exhibiting black spot parasites.

User	Watershed	2015	2016	2017	2018	2019
birds_bugs_botany	Northern Lake Erie	–	–	–	3	11
bkorol	Eastern Georgian Bay	–	1	–	4	7
	Eastern Lake Huron	–	–	–	–	1
oridgen10	Central Ottawa	–	–	–	–	1
	Lake Ontario and Niagara Peninsula	–	–	–	2	6
	Northern Lake Erie	–	–	–	2	–
	Potomac	–	–	–	–	1
pmk00001	French Broad-Holston	–	–	–	–	2
	Neosho	1	–	–	–	–
	Potomac	–	–	2	–	1
	Roanoke	1	–	–	–	1
	St. Croix	–	–	–	1	–
Upper Mississippi-Black-Root	–	–	–	1	–	
scottgibson	Lake Ontario and Niagara Peninsula	–	–	–	6	2
quiggifur	Apalachicola	–	–	–	5	–
	Lower Tennessee	–	–	2	–	–
reuve nm	Lake Ontario and Niagara Peninsula	–	–	–	3	–
	Northern Lake Erie	–	–	–	1	–
bradleyfishes	Central St. Lawrence	1	–	–	–	–
	Lake Ontario and Niagara Peninsula	–	–	–	1	–
	Santee	–	–	–	–	1
	Upper Pee Dee	–	–	1	–	–
dan_macneal	Northern Lake Erie	–	–	–	–	4
tortuga_rapido	Southern Lake Erie	–	–	4	–	–

better predictor than the location, East Georgian Bay watershed. When this user was removed from the dataset, the decision tree collapsed this node leaving splits based on if the data were from either the Northern Lake Erie watershed or the Lake Ontario and Niagara Peninsula watersheds or were not. Common name or group (stoneroller or chub) did not appear in the decision tree suggesting that predicting where signs of black spot were more prevalent was similar in each of the four groups

investigated. Similarly, year did not appear as a factor in deciding to classify observations into “infected” or not and thus not as important as the watershed of capture.

4. Discussion

Among 1383 fish photos a large percentage of fish from the adjacent East Georgian Bay, Lake Ontario – Niagara Peninsula, and Northern Lake Erie watersheds, together representing southern Ontario, Canada exhibited signs of black spot disease related pathogens. The overall frequency of occurrence of evidence for black spot metacercariae in southern Ontario (27% of 314 fish) is higher than the mean prevalence of 6.8% (median = 5.75%) for other watersheds with more than 10 fish observed in our study. Ecological, biological, and environmental reasons for the geographic pattern are difficult to prescribe without mining other data sources for water quality and land-use variables (i.e., Haas et al., 2018), or knowing more about the diversity of trematode species causing such characteristic black spots to appear, but herein I illustrate that the pattern exhibited is not likely due to user-related biases in data provided by community members.

The consistent geographic pattern noted in fish exhibiting signs of black spot disease does not appear to be simply due to the number of fish caught in the watershed. For example, none of the stonerollers caught in the Southern Lake Erie ($n = 35$), Little River (Texas; $n = 26$) or Middle Colorado River (Texas; $n = 26$) watersheds exhibited signs of the parasite, an observation supported by anecdotal evidence that few to no stonerollers are encountered with the characteristic black spots in Texas (personal communication: K. Mayes and G. Linam with Texas Parks and Wildlife Department, and A. Cohen with University of Texas: Austin). Similarly, none of the chubs observed in Virginia (Neuse River; $n = 18$) and Arkansas (Upper White River; $n = 15$) exhibited signs of infection. Southern Lake Erie remained a watershed that reported many observations for all species examined ($n = 114$ in total), but few ($n = 6$; 5.3%) exhibited evidence of black spot disease within the photos regardless of the fish species. As such, it seems unlikely that users are only posting photos of fish with spots out of a bucket of fish, or only catching fish exhibiting external signs of infection with metacercariae.

Admittedly, there is great potential for user bias when working with community-sourced data. When observations of fish with signs of black spot pathogens were summarized by the watershed of capture, as well as user, the number of observations classified as “infected” from a single user in Eastern Georgian Bay appeared large in comparison to the total observed in that watershed. Conditional inference tree analysis confirmed that classifying data by the user in this case was more accurate than by the East Georgian Bay watershed from which the fish were observed. This did not occur elsewhere in the inference tree, and thus

Table 4

Numbers of species caught and posted to iNaturalist in Southern Ontario watersheds by year and, in parentheses, the percentage of those fish that had visible signs of black spot infections.

	Blacknose Dace	Campostoma spp.	Creek Chub	Nocomis spp.
2015	1 (0)	–	2 (0)	1 (0)
Eastern Georgian Bay	–	–	1 (0)	–
Lake Ontario and Niagara Peninsula	1 (0)	–	1 (0)	1 (0)
2016	4 (50)	1 (0)	5 (20)	2 (0)
Eastern Georgian Bay	1 (100)	–	3 (33.3)	2 (0)
Lake Ontario and Niagara Peninsula	1 (0)	–	1 (0)	–
Northern Lake Erie	2 (50)	1 (0)	1 (0)	–
2017	6 (0)	–	14 (7.1)	7 (14.3)
Eastern Georgian Bay	–	–	2 (0)	2 (0)
Lake Ontario and Niagara Peninsula	6 (0)	–	11 (9.1)	4 (0)
Northern Lake Erie	–	–	1 (0)	1 (100)
2018	39 (17.9)	15 (33.3)	59 (15.3)	42 (40.5)
Eastern Georgian Bay	7 (14.3)	2 (50)	10 (0)	9 (55.6)
Lake Ontario and Niagara Peninsula	29 (17.2)	8 (12.5)	42 (21.4)	26 (30.8)
Northern Lake Erie	3 (33.3)	5 (60)	7 (0)	7 (57.1)
2019	33 (27)	14 (71.4)	59 (25.4)	10 (80)
Eastern Georgian Bay	5 (40)	–	12 (16.7)	4 (75)
Lake Ontario and Niagara Peninsula	23 (17.4)	1 (0)	33 (21.2)	2 (100)
Northern Lake Erie	5 (60)	13 (76.9)	14 (42.9)	4 (75)

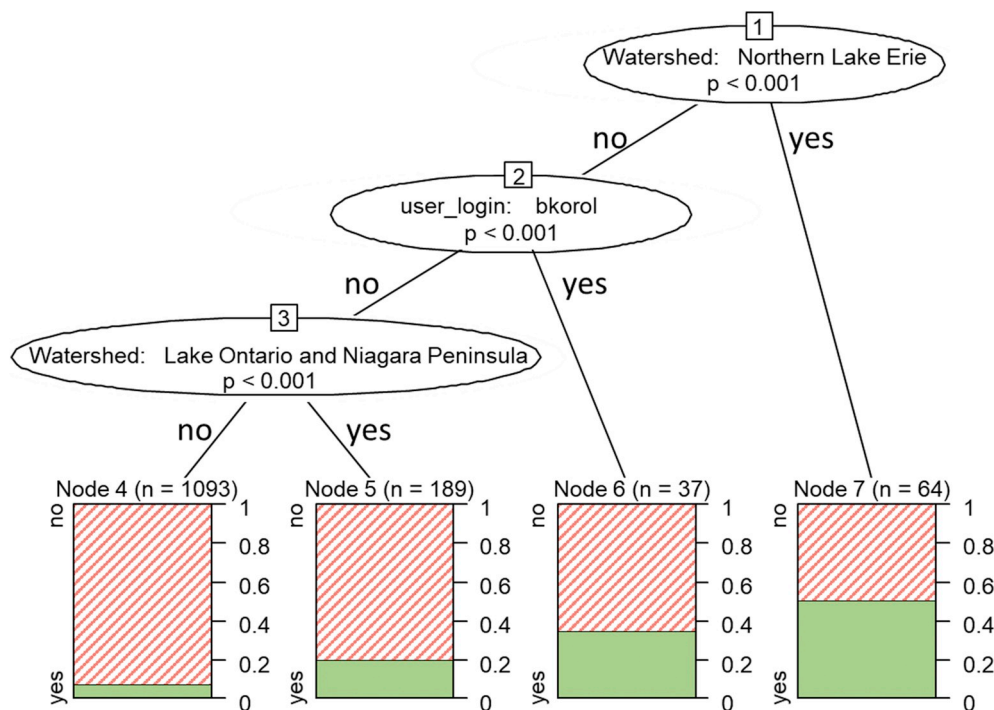


Fig. 3. If an observation of blacknose dace, creek chub, chubs (*Nocomis* spp.), or stone-rollers (*Campostoma* spp.) is reported on iNaturalist from the watersheds of Northern Lake Erie or Lake Ontario – Niagara Peninsula it is more likely to exhibit macroscopic signs of infection by metacercariae. Also a high percentage of the observations by user *bkorol* exhibited symptoms, all from Eastern Georgian Bay which is adjacent to the two other watersheds mentioned. Data were analyzed using conditional information trees in *r* using package *caret* to train and tune the model. Percentage of actual observations summarized in bar charts below each split that significantly described the data.

with an accuracy of 88.5% there was no evidence for other user-related biases in the data. Perhaps this user found fish in a stream with a particularly high infection rate compared to the other users in this watershed, conversely maybe others caught fish from areas with low infection rates, or just an anomaly. Regardless, the adjoining Lake Ontario – Niagara Peninsula and Northern Lake Erie watersheds did not have evidence of user-bias despite more users (54 and 17 respectively vs. 12 in East Georgian Bay) and had similar cross-species rates of being classified as “infected” with black spot pathogens. As such, there is not strong evidence that the main geographic pattern seen is due to any users over-reporting fish exhibiting black spots of any species included here.

There is also a possibility for an influence of year in the data,

whether due to natural cycles in parasite’s life history or due to differences in user activities (i.e., more users in watersheds with high infection rates in certain years). Data summarized by year for the watersheds of southern Ontario illustrates that the fish exhibiting signs of infection were not simply sampled in one bad year, the symptoms occur across several years. Data in Table 4 indicate that the frequency of encountering fish exhibiting signs of the disease increased each year within southern Ontario for each group of fish examined. Studies have suggested that warmer temperatures lead to higher rates of infections of black spot disease in salmonids (Cairns et al., 2005; Schaaf et al., 2017), perhaps climate change will increase infection rates. While the resulting inference tree did not indicate that year was an important predictor of black spot disease symptoms, observations from the same water body

over time would yield better insights into yearly differences, and the effects of climate change on variation in black spot infections.

Steedman (1991) concluded that wide, slow moving canals around Toronto led to higher prevalence of black spot disease, along with streams that were more impacted by urbanization. This thought was mirrored in anecdotal evidence of black spot disease rarely being seen in fish from cold, more pristine, headwater streams in Canada, compared to streams more impacted by human activities (personal communication; J. Barnucz from Fisheries and Oceans Canada, C. Chu and W. Wegman from Ontario Ministry of Natural Resources and Forestry, and R. Wilson from Lake Simcoe Region Conservation Authority). Watersheds used herein were large (HU6 or Sub-Basins) and thus encompass a variety of waterways and land uses. Blacknose dace, creek chub, and stonerollers are found throughout both heavily urbanized and lessor impacted areas (Iwanowicz et al., 2016; Wallace et al., 2013), whereas chubs appear to be comparatively less tolerant to urbanization than the other species included in this study (Helms et al., 2005), yet were found in similar watersheds. Unfortunately, observations reported from different waterbodies within each watershed, across different climates and degrees of urbanization do not lend well to an assessment of how urbanization affects black spot parasite populations. A more robust sampling protocol across urbanization gradients within a stream could offer better insights into how urbanization influences infection rates than the opportunistic observations included in this study.

As fish orientation in photographs is not standardized in iNaturalist, a judgement of the degree or intensity of infection (abundance of parasites) for each individual fish was not attempted. Whereas damage caused by one neascus-type trematode is negligible, greater infection numbers may be detrimental due to high disease or predation mortality (Wisenden et al., 2012). However, most photos show only one side of the fish and sometimes even that was partially obscured (i.e., by a hand holding the fish), so only presence or absence of signs of black spot pathogens was recorded. It would be helpful if users took care in the future to photograph fish on both sides, clear of debris, unobstructed, and preferably in a clear fish viewer.

It is important to note that other fish species are susceptible to black spot related metacercaria that were not reported on, and that black spot disease has been seen throughout the U.S. Others have seen high prevalence of the parasites in suckers (*Catostoma* spp.), darters (*Etheostoma* spp.), shiner (*Notropis* spp.), studfish (*Fundulus catenatus*) and other Cyprinidae spp. across much of Arkansas (McAllister et al., 2016, 2013). Steelhead trout (*Oncorhynchus mykiss*) in California appeared to be infected at higher rates when temperatures rose above 23 °C (Schaaf et al., 2017). Mellen (2001) saw black spot parasites in 33% of creek chub and blacknose dace, 35% of central stoneroller (*Camptostoma anomalum*), and < 11% of chubs (*Notropis* spp.) found in western Iowa. Quist et al. (2007) reported low prevalence of black spot in creek chub from Wyoming, but nearly 100% of the suckers and ~80% of chubs had black spot pathogens visible. Prevalence of neascus-type trematodes in central stonerollers was consistently different between two streams in Tennessee, attributed to abiotic factors (Ferrara and Cook, 1998). Chapman et al. (2015) found that around 30% of pumpkinseed (*Lepomis gibbosus*) near Cornwall Ontario had black spot parasites whereas Cone and Anderson (1977) examined pumpkinseed in Ryan Lake, Ontario, and found that the prevalence was at or near 100% for age-1 and older fish. Nearly 60% of fathead minnows (*Pimephales promelas*) contained black spot parasites in a lake in Minnesota (Wisenden et al., 2012). For mosquitofish (*Gambusia affinis*) a prevalence of >10% was noted in central Texas (Tobler and Schlupp, 2007). While not an exhaustive list, the above series of summaries does not lead one to a strong conclusion that there is a consistent geographic pattern to the presence or prevalence of black spot pathogens in any group of fish species. Herein, I included data on just four groups of fish species as they offer enough evidence to make a compelling story that data volunteered by community members can offer unique insights into parasite ecology as well as distributions for species, including pathogens, which have received

little attention.

Metacercaria of several different species induce the characteristic melanocytes giving black spot disease its moniker. As such, it is likely that trematode species differ in infectivity and geographic range, as well as having different responses to environmental factors which may be the underlying causes to the geographic patterns described in this study. Whereas studies into the prevalence of black spot disease typically microscopically inspect metacercaria for species confirmation (McAllister et al., 2013; Quist et al., 2007), or some simply use tactile confirmation of black nodules vs. natural coloration (Schaaf et al., 2017), these steps were unavailable for this study and only visual classification into “infected” vs. not was available, similar to Wisenden et al. (2012). Chandler (1951) noted that all black spots examined in yellow perch from a Minnesota lake contained a metacercaria of neascus-type trematodes. The size, shape, color, abundance, and seemingly random distribution of the characteristic black spots shown in Fig. 1 are unlikely to occur due to non-neascus-type etiologies (i.e., cuts, scrapes, punctures, natural color variations), those wishing to study other diseases in photograph databases are likely to need more ground-truthing causative agent's identification.

Data from iNaturalist are not currently conducive to directly studying distribution of a species directly as observations depend on the number of users and their interests. For example, the number of fish observations in a watershed is more representative of the number and angling skills of iNaturalist users looking for stream fish in each watershed than the actual abundance of the species. As such, direct observations of species tend to be skewed towards urban areas where they may not be as abundant as the iNaturalist data suggests. However, when the subject of study (i.e., a visible disease) is incidental alongside the main organism in the observation, such data can be treated more scientifically. A similar concept, Rowley et al. (2019) reviewed 892 photos of Australian amphibians on iNaturalist and saw that 6% were being handled without gloves, leading to disease implications and the potential to increase outreach efforts on proper handling methods. As such, we believe this study represents a novel means of exploring databases such as iNaturalist, ideally encouraging others to creatively utilize such large datasets.

In conclusion, I note that for two fish species, and across two other genera of fish, evidence of black spot parasites were exhibited in a higher percentage of observations from southern Ontario than other areas the fish were caught in. Causes for such a consistent geographic pattern are likely due to a combination of biological, ecological, and environmental factors rather than potential biases related to the use of community-member provided data. It is important to note that presence of parasites does not necessarily equate to poor ecosystem health, and instead diverse parasite communities are thought to occur in healthy ecosystems (Hudson et al., 2006; Marcogliese, 2005). For example, restoration efforts lead to increased prevalence of trematodes in Californian marsh, thought to be due to increased abundances of piscivorous birds (Huspeni and Lafferty, 2004). To my knowledge this is the first exploration of a community-member populated photo database for geographic patterns of symptoms of a disease. More importantly these findings illustrate that research endeavors can arise from these volunteer-populated databases and that the databases should not be discounted (Cohn, 2008). Continued data collection by community members and subsequent correlation with environmental datasets may help elucidate causative agents to the observed geographic pattern.

Conflicts of interest

The author states that there are no conflict of interests.

Data availability statement

All observations used were obtained from and are available from the iNaturalist.org website. Data on the presence/absence of black spot in

the observations available from the author upon request. Watershed polygons originated from the Commission for Environmental Cooperation (CEC, 2011) and were obtained from USGS ScienceBase (<https://www.sciencebase.gov/catalog/item/4fb697b2e4b03ad19d64b47f>).

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