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First records of Finescale Dace (*Chrosomus neogaeus*) in Newfoundland and Labrador, Canada

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Abstract
The island of Newfoundland has no official record of cyprinid fishes. Here, we report the discovery of a minnow, Finescale Dace (*Chromous neogaeus*) from four ponds located in a first order tributary of the Exploits River, in central Newfoundland. This finding represents the first record of the species in the province. The location where the species was found is in a localized, central portion of insular Newfoundland, therefore, the most parsimonious explanation for this new record is that it was an illegal, intentional introduction. Such introductions in other provinces have occurred by anglers who felt it would serve as a forage fish for other species. The consequences of this introduction to native species are unknown; however, the dace's local abundance, foraging behaviour, and reproductive capacity are discussed in terms of the interspecific competition with native species.

Key words: Finescale Dace; *Chrosomus*; Goldfish; intentional introduction; interspecific competition; exotic species

Introduction

The province of Newfoundland and Labrador has a very low rate of introduction for exotic fish (van Zyll de Jong *et al*. 2004). There are 28 native freshwater fish species in Newfoundland and Labrador; 15 species can be found in insular Newfoundland with an additional 13 species occurring in Labrador (van Zyll de Jong *et al*. 2004; Table 1). For the island portion of the province there have been five attempts at species introductions. These introductions were all salmonids including Brown Trout (*Salmo trutta*), Rainbow Trout (*Onchorhynchus mykiss*), Lake Whitefish (*Coregonus clupeaformis*), Lake Trout (*Salvelinus namaycush*), and Pink Salmon (*Oncorhynchus gorbuscha*; Table 1). However, of these five introductions only the first two succeeded. These species were introduced by government agencies intending to use stocking programs to enhance freshwater fisheries (Scott and Crossman 1964; Hustins 2007).

The only other account of a species introduction documented for the province is Goldfish (*Carassius auratus*). This species has been reported in three locations, but as yet, has not been substantiated by a government agency. The three alleged locations include Mundy Pond (47.55152°N, 52.73891°W) centrally located in the city of St. John's, a small pond near the town of Heart's Delight (47.78691°N, 53.46752°W), and Janes Pond (47.16617°N, 55.16229°W) located in the town of Marystown. All of these locations are in proximity to urban areas, suggesting the likelihood of intentional release of household aquaria pets. In Labrador, there are no records of exotic freshwater fish.

On 4 June 2010 the provincial department of Environment and Conservation received a request to identify an unusual fish that had allegedly been angled by a sport fisher in central Newfoundland. The examination of a photograph suggested the unknown species was a minnow from Cyprinidae family. The fish was reportedly angled from a pond on a tributary of the Exploits River. A field trip was subsequently planned to verify the report or determine the extent of its presence. Herein we report on the occurrence of an exotic invasive minnow species existing in four ponds of a first order tributary of the Exploits River watershed, central Newfoundland.

Methods

Sample collection

To verify the species present and the extent of their distribution two site visits occurred. The first occurred 4–16 June 2012 and the second 9–17 June 2014. During the latter trip the number of sites

TABLE 1. A list of native and introduced freshwater fish species of Newfoundland and Labrador, Canada (van Zyll de Jong *et al.* 2004).

Common name	Scientific name	Newfoundland	Labrador
American Eel	*Anguilla rostrata*	+	+
Fourspine Stickleback	*Apeltes quadracus*	+	−
Threespine Stickleback	*Gasterosteus aculeatus*	+	+
Blackspotted Stickleback	*Gasterosteus wheatlandi*	+	−
Ninespine Stickleback	*Pungitius pungitius*	+	+
Atlantic Salmon	*Salmo salar*	+	+
Brook Trout	*Salvelinus fontinalis*	+	+
Arctic Char	*Salvelinus alpinus*	+	+
Lake Trout	*Salvelinus namaycush*	−	+
Lake Whitefish	*Coregonus clupeaformis*	−	+
Round Whitefish	*Prosopium cylindraceum*	−	+
Rainbow Smelt	*Osmerus mordax*	+	+
Sea Lamprey	*Petromyzon marinus*	+	+
Alewife	*Alosa pseudoharengus*	+	+
American Shad	*Alosa sapidissima*	+	+
Atlantic Tomcod	*Microgadus tomcod*	+	+
Banded Killifish	*Fundulus diaphanus*	+	−
Mummichog	*Fundulus heteroclitus*	+	−
Northern Pike	*Esox lucius*	−	+
Lake Chub	*Couesius plumbeus*	−	+
Longnose Dace	*Rhinichthys cataractae*	−	+
Northern Pearl Dace	*Margariscus nachtriebi*	−	+
Longnose Sucker	*Catostomus catostomus*	−	+
White Sucker	*Catostomus commersonii*	−	+
Burbot	*Lota lota*	−	+
Mottled Sculpin	*Cottus bairdii*	−	+
Slimy Sculpin	*Cottus cognatus*	−	+
Logperch	*Percina caprodes*	−	+
Rainbow Trout*	*Onchorhynchus mykiss*	+	−
Brown Trout*	*Salmo trutta*	+	−
Goldfish*	*Carassius auratus auratus*	+	−

*Introduced species.

searched was expanded. Initial samples were taken from four headwater lakes located on the south side of the Exploits River near the town of Bishop's Falls. The lakes are located on a first order tributary (Jumpers Brook 49.02667°N, 55.40216°W: Tributary 1; Figure 1). Jumpers Brook flows into the main stem of the Exploits River (49.02857°N, 55.40164°W), entering the Exploits ~8 km upstream from the Atlantic Ocean (in Exploits Bay). Three of the sampled ponds (Mina 1, Mina 2, and Mina 3) were in close proximity (<1 km) to each other and were both small (surface area <0.10 km^2) and shallow (<2 m depth). The fourth lake, Mina 4 was larger (surface area = 0.35 km^2) and deeper (>4 m depth). Mina 4 was ~2 km upstream from Mina 3. The substrate in each pond was mud with scattered rubble and cobble with emergent grasses surrounding the shorelines. The water was dark in colour, the product of humic conditions. During the 2012 sampling, surface temperatures ranged from 8°C to 10°C. In 2014 the temperatures were between 11°C and 20°C.

During the first sampling event, in 2012, both fyke (FYKE-001-06, 10 mm mesh, Shandong, China) and gill nets (Miller Nets, Memphis, Tennessee, USA) were used. Gill nets were composed of nylon monofilament and each set consisted of two stretch mesh sizes measuring 1.91 cm and 2.54 cm, with the smallest mesh being set closest to the shoreline. Three gill nets were set in Mina 1, two in Mina 2 and 3, and four sets were placed in Mina 4. All gill nets were set perpendicular to the shoreline and were set overnight, for an average of 12 h. Fyke (trap) net exterior netting consisted of 1.91 cm stretch mesh size with front hoop of 1 m through to four cylindrical interior hoops with a tied trap end. These nets were set with a 9.8 m mesh lead extending from the front hoop perpendicular to the shoreline. This lead net also consisted of 1.91 cm stretch mesh size. Three small fyke nets were set in the littoral area of Mina 2. All captured native species were retained for measurement and stomach examinations in the laboratory. A subset of unidentified minnow species were retained and preserved in 95% ethanol.

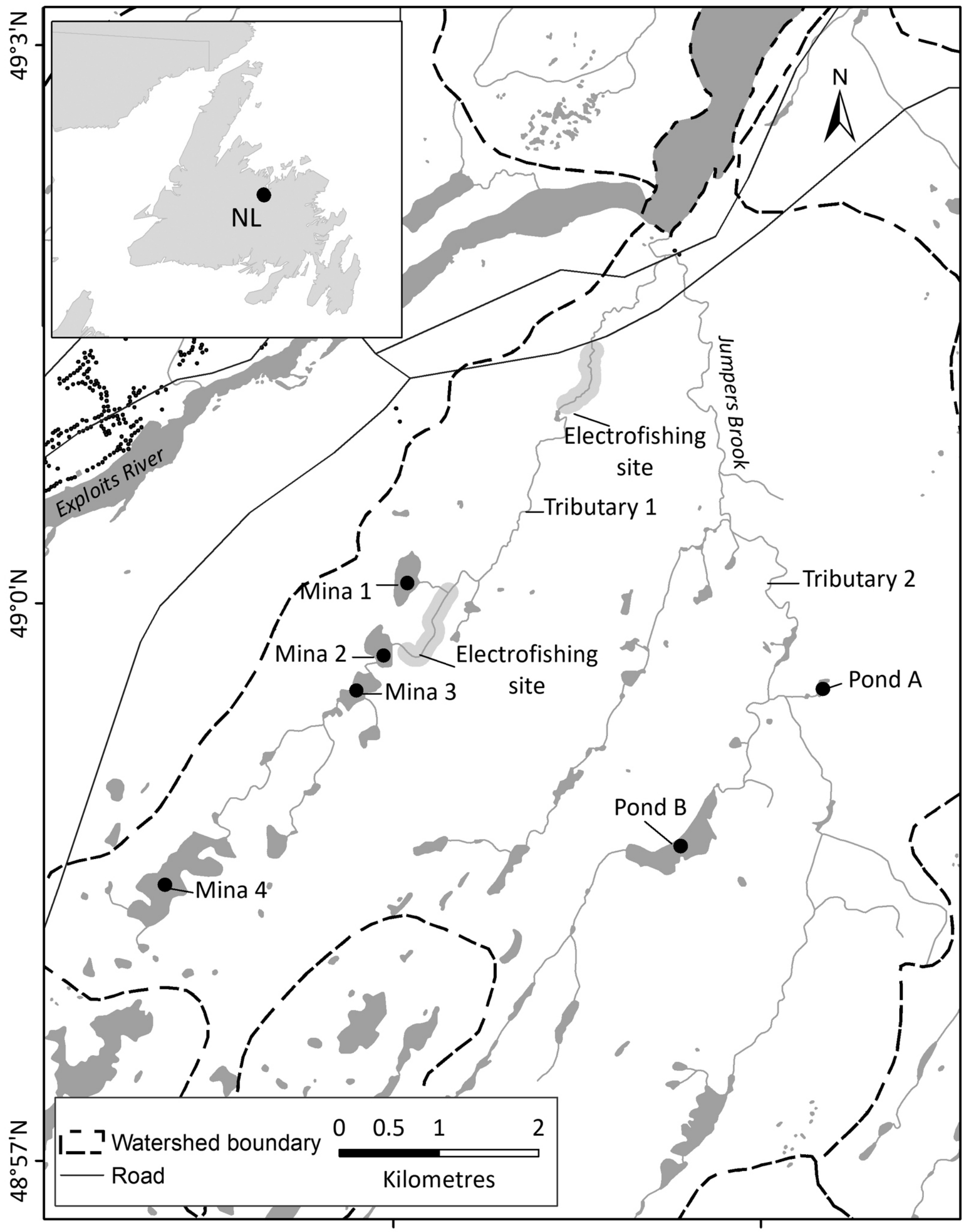

FIGURE 1. The four ponds (Mina 1, 2, 3, and 4) on Tributary 1 from which Finescale Dace (*Chrosomus neogaeus*) was sampled, central Newfoundland. Also shown are the locations of Tributary 2, Ponds A and B, and the locations of the electrofishing stations.

In 2014, sampling was repeated on each of the four ponds originally sampled in 2012. However, instead of gill and fyke nets, minnow pots were used as they have proven to be very effective when sampling dace (He and Lodge 1990). The minnow pots (Gees Feets G-40 Minnow trap, Tackle Factory, Fillmore New York, USA) were constructed of galvanized steel wire with length 42 cm, diameter 23 cm, and square mesh size of 6 mm. Thirty-nine overnight pot sets were placed in ponds Mina 1, 2, 3, and 4 (Table 2). A

TABLE 2. Year and location of sampling, gear type, and numbers set (*n*). Frequency of fish sampled per species for all ponds 4–16 June 2012 and 9–17 June 2014 in central Newfoundland. All sets were overnight.

Year	Location	Gear Type (*n*)	Brook Trout (*Salvelinus fontinalis*)	Atlantic Salmon (*Salmo salar*)	Threespine Stickleback (*Gasterosteus aculeatus*)	Finescale Dace (*Chrosomus neogaeus*)
2012	Mina 1	Gill (3)	29	1	2	10
	Mina 2	Gill (2)	10	9	1	1
	Mina 2	Fyke (3)	7	1	3	—
	Mina 3	Gill (2)	—	—	—	—
	Mina 4	Gill (4)	9	—	—	3
2014	Mina 1	Pot (16)	2	—	26	391
	Mina 2	Pot (9)	—	—	19	24
	Mina 3	Pot (6)	—	—	43	2
	Mina 4	Pot (8)	—	—	15	110
	Pond A	Pot (4)	—	—	—	—
	Pond B	Pot (3)	6	—	29	—

sub-sample of 20 unidentified minnow species were retained, preserved in 95% ethanol, and sent to the Department of Biology at Dalhousie University for genetic-based identification. The sub-sample was a random selection of 10 male and 10 female fish.

To determine if the dace were isolated to just the four ponds or were present in other areas of Jumpers Brook, the lower (1.5 km) and upper (1 km) reaches of Tributary 1 (Figure 1) were sampled by electrofishing. Two small ponds located in another first order tributary were also sampled (Tributary 2; Figure 1). In Tributary 2, four pots were set in Pond A and three pots were placed in Pond B.

Morphometric and subsequent genetic analysis

External morphological measures were recorded for 200 dace chosen from the 2014 collection (Table 3). With the exception of fork length, the morphological characters measured followed those described by Hubbs and Lagler (1958) and Doeringsfeld *et al.* (2004) which included the predorsal length, pectoral fin position, head length, maximum body width, maximum body depth, caudal peduncle depth, interorbital width, gape width, and the upper jaw length.

Species identification using the morphometric data was attempted but proved difficult for several reasons. Given the morphological variability among the sampled dace it was determined that it could be one, or a combination of, three possible species, each sharing very similar physical characteristics. The three species included Northern Redbelly Dace (*Chrosomus eos* (Cope, 1862)), Finescale Dace (*Chrosomus neogaeus* (Cope, 1867)), or their hybrid *Chrosomus eos-neogaeus*. Subsequently, genetic analysis was used for determining species. Nevertheless, this did not remove all the difficulty in identification.

Standard DNA barcoding/sequencing methods using the mitochondrial CO1 gene would not be suf-

TABLE 3. Variation in morphological characters measured from 200 *Chrosomus* sp. collected 9–17 June 2014 from ponds in the Jumpers Brook watershed, central Newfoundland. Measurements as described by Hubbs and Lagler (1958) and Doeringsfeld *et al.* (2004): fork length (FL), predorsal length (PDL), pectoral fin position (PFP), head length (HDL), maximum body width (BYW), maximum body depth (BYD), caudal peduncle depth (CPD), interorbital width (IOW), gape width (GPW), upper jaw length (UJL).

Character	Range	95% CI	Mean	SE
FL	39.00–93.00	64.7–68.7	66.69	0.998
TBM	0.71–10.23	3.64–4.26	3.95	0.157
PDL	18.00–48.10	32.80–34.86	33.83	0.520
PFP	6.30–20.70	13.67–14.50	14.08	0.210
HDL	7.90–21.23	13.81–14.65	14.23	0.214
BYW	4.40–15.10	8.28– 8.92	8.59	0.162
BYD	7.50–20.80	13.35–14.25	13.80	0.228
CPD	0.30–3.60	1.27–1.46	1.37	0.046
IOW	2.50–8.50	5.58–5.97	5.78	0.098
GPW	3.90–13.20	8.01–8.55	8.28	0.136
UJL	1.70–8.90	4.64–4.95	4.79	0.079

ficient for all necessary species identifications in this case. Hybrid individuals (*C. eos-neogaeus*) share mitochondrial DNA with their maternal parent, most commonly *C. neogaeus* (Binet and Angers 2005) and these standard identification techniques would not allow us to distinguish these two complexes. As a result, a method of species identification based on nuclear DNA, in addition to mitochondrial DNA, was required.

Caudal fin tissues were digested with proteinase K (Bio Basic Inc., Markham, Ontario, Canada) for ~8 h at 50°C. DNA was extracted from these tissue digests following a glassmilk approach modified from Elphinstone *et al.* (2003) for use with a Multiprobe II liquid handling system (PerkinElmer, Waltham, Massachusetts, USA). The resulting DNA was examined for quality and quantity by means of gel electrophoresis using a 1% agarose gel.

To perform the species identification, we followed the recommendations found in Binet and Angers (2005) in which the GH and the PEG1/MEST nuclear loci were amplified using polymerase chain reaction (PCR). The resulting PCR products were visualized on a 2% agarose gel and examined for band combination and size to detect the presence of *C. neogaeus*, *C. eos*, or their hybrid.

Five complete *Chrosomus* sp. from the 2012 collection were archived at the Royal Ontario Museum (accession number: ROM 104124). Additionally, 20 caudal fin tissues, used for genetics-based identification, were transferred to The Rooms Corporation of Newfoundland and Labrador, Provincial Museum Division (accession number: NFM T-2018-63). All specimens were preserved in 95% ethanol.

Results

Species account

In 2012, native fish species captured included Brook Trout (*Salvelinus fontinalis*), Atlantic Salmon (*Salmo salar*), and Threespine Stickleback (*Gasterosteus aculeatus*; Table 2). In addition, 14 dace were also sampled from three of the four ponds surveyed (Mina 1, 2, and 4; Table 2). Stomach examinations revealed that one Brook Trout, from Mina 1, contained approximately 20 partially digested young-of-the-year dace (Figure 2a).

In 2014, 527 dace were sampled from all four lakes, for an average capture of 13.5 fish per pot. Figure 2b shows three representative specimens from the collection. In Tributary 2 we did not capture any dace in either Ponds A or B, nor did we sample any dace when electrofishing the lower and upper reaches of Tributary 1. Only native species Brook Trout, American Eel (*Anguilla rostrata*), Atlantic Salmon, and Threespine Stickleback were sampled.

Genetic results

Of the 20 selected samples, 16 yielded sufficient DNA quantity and quality for species identification. The remaining four were excluded due to DNA quality concerns. Our examination of the amplifications of the GH locus showed band size uniformity among all individuals, with a band size of approximately 250 bp. This particular fragment/band size is indicative of *C. neogaeus* (Binet and Angers 2005). A similar result was observed for the PEG1/MEST amplifications: band size uniformity of approximately 177 bp. Again, this particular band size is indicative of *C. neogaeus* (Binet and Angers 2005). There was no

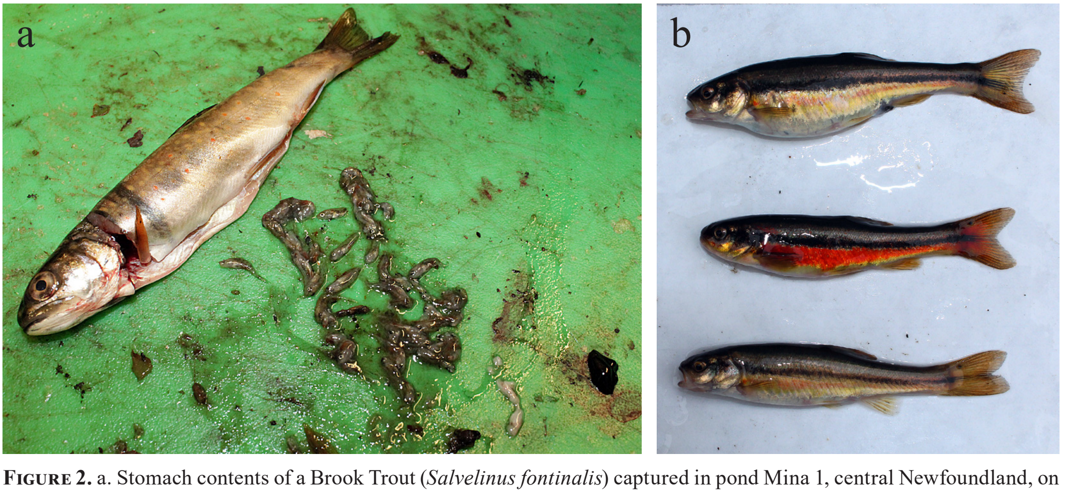

FIGURE 2. a. Stomach contents of a Brook Trout (*Salvelinus fontinalis*) captured in pond Mina 1, central Newfoundland, on 7 June 2012, showing approximately 20 partially decomposed immature cyprinid species. b. Finescale Dace (*Chrosomus neogaeus*) sampled from pond Mina 2, central Newfoundland, on 14 June 2014 Photo: a. Robert Perry. Photo: b. Donald Keefe.

evidence of any other band size or combination in any of the 16 individuals tested at either of these two nuclear DNA loci, indicating the absence of both hybrid *C. eos-neogaeus* and *C. eos* individuals.

Discussion

The record of Finescale Dace in four headwater ponds in the Exploits River watershed represents the first report of an introduced Leuciscinae species for insular Newfoundland. The species complement for the island portion of the province contains only euryhaline (saltwater tolerant) species (Scott and Crossman 1964; van Zyll de Jong *et al*. 2004) and Finescale Dace is not considered euryhaline. Therefore, it is unlikely that the species colonized the area naturally and indicates that an exotic introduction has occurred. The known distribution of Finescale Dace extends across the southern and northwest parts of Canada (Scott and Crossman 1998). In the east, its distribution reaches New Brunswick and Nova Scotia while in the United States it can be found in the Mississippi and Missouri river drainages (Stasiak 1980; Page and Burr 1991). The origin for the Jumpers Brook population is uncertain.

Because young-of-the-year minnows were found during stomach examination of Brook Trout it is evidence that the nonindigenous species is established. The habitat preferences for Finescale Dace include acidic beaver ponds, boggy waters, and small lakes with bottom detritus and silt (Stasiak and Cunnigham 2006). The locations in which we discovered Finescale Dace fit the habitat requirements for the species (Mee and Rowe 2010).

In accordance with our 2014 field sampling, we did not find evidence for the presence of Finescale Dace in either Tributary 1 or in the headwater ponds of Tributary 2. It would appear that the presence of the dace is currently limited to the four headwater ponds of Tributary 1. We believe the containment is owing to the presence of beaver dams in ponds Mina 1 and Mina 2 which limit the outflow of water and impedes fish movement. The outflow from Mina 2 is downstream from Mina 3 and 4 and therefore controls the flow of water from these two ponds.

If the intent of the introduction was to provide a food source for both Brook Trout and juvenile Atlantic Salmon, it may have been short-sighted. Although we did find juvenile dace in the stomach of an adult Brook Trout, it is unclear how additional competition for food sources will affect native fish populations into the future. Finescale Dace are sight feeding, carnivorous predators and have been documented as eating a wide variety of food items including aquatic insects, vegetation, and molluscs (Stasiak and Cunnigham 2006; Mee *et al*. 2013). For example, adult fish have been documented eating midges (Diptera), beetles (Coleoptera), caddisflies (Trichoptera), mayflies (Ephemeroptera), and bugs (Hemiptera; Stasiak 1972). Native species such as Brook Trout, Atlantic Salmon, and Threespine Stickleback depend upon micro and macro invertebrates, particularly at juvenile stages (Scott and Crossman 1998). Further, these types of minnows have a large reproductive capacity. It has been reported that Northern Redbelly Dace can produce 316 000 fish per ha of surface area in a single season (Cooper 1935). Finescale Dace also has a substantial reproductive capability. For example; a 70 mm female may produce 2600 eggs (Stasiak 2011). Given the potential for these types of cyprinids to have a large reproductive capacity, it is probable that the presence of Finescale Dace will lead to interspecific resource competition with native species.

Competition among piscivores and planktivores can have a large influence on rates of primary production in an aquatic ecosystem (Carpenter *et al*. 1985; Carpenter and Kitchell 1988). Following the introduction of Eurasian Minnow (*Phoxinus phoxinus*) into a Norwegian subalpine lake, Øvre Heimdalsvatn, researchers witnessed major changes to both the structure and composition of macroinvertebrate benthos (Naestad and Brittain 2010). These changes led to negative effects on the native Brown Trout population, including reduced annual recruitment and growth rates. Researchers attributed the changes to competition for food (Borgstrom *et al*. 2010).

There may also be a direct effect on native species abundance, as these types of predatory minnows have been documented eating the eggs and young of other species. A dissected large male Finescale Dace was reported having its intestinal tract full of fish eggs (presumed to be the eggs of the Northern Redbelly Dace; Stasiak 1972). Becker (1983) reported that dace will eat guppies in an aquarium setting. Furthermore, it has been reported that Northern Redbelly Dace, a species whose dietary preferences overlap with the Finescale Dace (Cochran *et al.* 1988), would eat Smallmouth Bass (*Micropterus dolomieu*) fry (Scott and Crossman 1999). Thus, it is possible that fry of Brook Trout and stickleback could also provide a food source for Finescale Dace. Further work is required to determine the short and long-term impacts of this introduction on native fauna.

The discovery of Finescale Dace in this particular watershed is disconcerting. The Exploits River represents one of the largest Atlantic Salmon producing rivers in North America (Pinfold 2011). The spread of this stenohaline species throughout the Exploits watershed may be limited by the presence of the Grand Falls Hydro-electric facility. The facility serves as a major barrier to fish passage and is located just upstream from where the tributary containing dace enters the main branch of the Exploits River.

The origin of Finescale Dace found in the four headwater ponds is uncertain. However, there are two possible explanations. The species current documented range limit in eastern Canada are the provinces of New Brunswick and Nova Scotia (Stasiak 1980; Page and Burr 1991). Therefore, it is possible that anglers imported the species as an illegal form of bait from the Maritime region. However, this explanation seems improbable given that these ponds are small in size and not known as a favoured area for recreational fishing. Another possible explanation may be that minnows kept in aquaria by area residents were released. Local residents interviewed regarding this possibility suggested that an unknown species of minnow had been housed in close proximity to the headwater ponds. Speculation by those interviewed was that minnows were imported from Labrador. Although interesting, this idea is unsubstantiated.

The presence of Finescale Dace in Labrador (even if remote) is an intriguing possibility. The present range distribution for Finescale Dace does not extend into Labrador and the only known species of dace existing in the province is Northern Pearl Dace (*Margariscus nachtriebi*). Thus, one may speculate that an undiscovered species is present or, alternatively, the current identification for the species in Labrador maybe an error. Further genetic work is required to investigate these possibilities.

Our findings suggest these minnow species are readily trapped and therefore could easily be moved as bait to other regions of the province. However, it is illegal in Newfoundland and Labrador to transport live fish or to use live fish as bait in inland waters. It is hoped that these regulatory restrictions will serve as a deterrent to the spread of the species. Additionally, to limit the spread of Finescale Dace, we recommend the launch of a public education and awareness program pertaining to the existence of these species in the province. Such an initiative would be beneficial and low cost for regulatory authorities. Through this effort, it is hoped that further illegal introductions or transfers could be curtailed. Additionally, the four ponds where these dace were found were small and therefore we recommend some consideration should be given to an eradication program to halt their spread.

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