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Taxonomic resolution of the North American invasive species of the genus *Bythotrephes* Leydig, 1860 (Crustacea: Cladocera: Cercopagididae)

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Abstract

The invasive species of the genus *Bythotrephes* introduced in the North American lakes in 1970s-early 1980s was named variously either *B. cederstroemi* or *B. longimanus*. The investigation of individuals of the genus from 15 Canadian lakes has allowed us to identify all of them as *B. cederströmii* Schödler, based on the detailed taxonomic redescription of the Eurasian representatives of the species. The available documented data (figures and photographs) on *Bythotrephes* from other North American lakes, made it possible to recognize their identity with this species as well. The possible introduction of interspecific hybrids of the genus *Bythotrephes* in the North American lakes has not been confirmed.

Key words: *Bythotrephes cederströmii,* identification, description, North American lakes

Introduction

The representatives of the invasive genus *Bythotrephes* Leydig, 1860 were first detected, being mostly recorded as a part of fish gut content, in the zooplanktonic communities of the Laurentian Great Lakes in early to mid-1980s (Bur *et al*. 1986; Lange & Cap 1986; Evans 1988; Cullis & Johnson 1988; Johannsson *et al*. 1991). However, according to lake sediment studies they might have been introduced in the lake communities earlier (Branstrator *et al*. 2017) albeit in a low number making them below detection by lake sampling efforts. Subsequent to colonization of the Great Lakes, *Bythotrephes* widely invaded the neighboring inland waters of the United States and Canada (Yan *et al*. 1992, 2011; Kerfoot *et al*. 2016; Jansen *et al.* 2017) and continues to expand its distribution, likely inadvertently dispersed by recreational boaters (Kerfoot *et al*. 2011, Kelly *et al*. 2013).

The appearance of *Bythotrephes* in North America caused an explosion of interest in these crustaceans. Their impact on inland water communities in their invaded range is great, resulting in reduced zooplankton richness and abundance, primarily associated with predation on cladocerans (Yan *et al*. 2011; Kerfoot *et al*. 2016). Many scientific publications are devoted to the description of its biology. For this reason, the North American *Bythotrephes* could be fairly called "the world's best studied invasive zooplankter" (Bollens *et al*. 2002; Yan *et al*. 2011).

It is paradoxical that, despite being deeply investigated ecologically, the taxonomic status of the North American *Bythotrephes* remains poorly understood. It was initially identified as *B. cederströmii* Schödler, 1877 ("*B. cederstroemii",* "*B. cederstroemi*") (see Bur *et al.* 1986; Lange & Cap 1986; Evans 1988; Cullis & Johnson 1988; Sprules *et al*. 1990; Johannsson *et al*. 1991, etc.) but some researchers doubted the correctness of the species designation (e.g., Lehman 1987; Evans 1988; Sprules *et al*. 1990). They noted the considerable intra-and interpopulational morphological variability of *Bythotrephes* and identified them variously as either different species, *B. longimanus* Leydig, 1860 and *B. cederströmii,* or different morphotypes of one species. The latter opinion has prevailed largely as a consequence of the results of European investigators who tended to treat the genus *Bythotrephes* as monotypic, being represented by only one quite variable species (Lieder 1958; Zozulya & Mordukhai-Boltovskoi 1977; Mordukhai-Boltovskoi & Rivier 1987). Subsequent research, supported by genetic studies, followed the same line and accepted "*B. longimanus*", which is both morphologically and genetically variable and widely distributed, as the only species of the genus (Berg & Garton 1994; Martin & Cash-Clark 1995; Grigorovich *et al.* 1998; MacIsaac *et al*. 2000; Therriault *et al*. 2002; Colautti *et al*. 2005; Yan *et al*. 2011; Kerfoot *et al*. 2016).

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All these researchers, however, were misled by the inadequate studies of the European representatives of the genus, whose species richness has proved to be higher than was thought initially. The higher taxonomic diversity of *Bythotrephes* was postulated by Lilljeborg (1901) and especially Ischreyt (1934, 1936, 1939) but their considerations were later either abandoned or rejected. This was mainly due to inadequate investigation of highly morphologically variable and poorly studied representatives of *B. cederströmii* or yet undescribed interspecific hybrid forms which were incorrectly collectively considered as members of the variable species, "*B. longimanus*" (Zozulya & Mordukhai-Boltovskoi 1977; Mordukhai-Boltovskoi & Rivier 1987; Grigorovich *et al*. 1998; Rivier 1998; Rivier & Grigorovich 1999). The existence of hybrids of *Bythotrephes* confused researchers who attempted to resolve the taxonomic structure of the genus using either morphological or genetic methods. Studies of morphologically highly variable hybrids or amalgam of forms, including specimens of different undistinguished species and hybrids, have inevitably resulted in the incorrect conclusion that only one variable species of the genus existed.

Later, Litvinchuk (2002, 2007) and Litvinchuk & Litvinchuk (2016) provided morphometric and allozymic analyses of numerous specimens of *Bythotrephes,* mostly from populations in the north-western and central parts of European Russia. As a result, the validity of five species of the genus, "*B. longimanus* Leydig, 1860*, B. cederströmii* Schoedler, 1863*, B. crassicaudus* Lilljeborg, 1890, *B. brevimanus* Lilljeborg, 1901, and *B. transcaucasicus* Behning, 1941", was confirmed along with interspecific hybrid forms. Subsequently, these species and hybrid forms have been intensively investigated, revised and redescribed in detail (Korovchinsky 2015, 2016, 2018, 2019), revising the long-used simplified taxonomic scheme of the group.

Based on results of the aforementioned recent taxonomic publications, we investigated the morphology of *Bythotrephes* from 15 Canadian lakes to reassess the taxonomic status of the North American members of the genus.

Material and methods

Study area. Sample specimens from Ontario, Canada were collected as part of the Ontario Ministry of Natural Resources and Forestry's (OMNRF) Broad-scale Fish Community Monitoring (BsM) Program (Sandstrom *et al.* 2015). Lake locations range from northwestern to southcentral Ontario (Figure 1) in the Boreal Shield ecoregion. These lakes support fish with economic value, including lake trout (*Salvelinus namaycush* (Walbaum, 1792)), walleye (*Sander vitreus* (Mitchill, 1818)), and brook trout (*Salvelinus fontinalis* (Mitchill, 1814)). Most of the sampled lakes are large (median surface area = 1806 ha), low mineralized (median conductivity = 45 uS/cm), and nutrient-poor (median total phosphorus = 8.6 ug/L) (Table 1). In addition, several specimens were collected from Lake Winnipeg, a large, shallow, eutrophic lake in Manitoba (Table 1).

Specimens were examined using the dissecting microscope Lomo MBS and compound microscope Olympus BX-41 with *camera lucida.* Body measurements were made according to the scheme presented in Korovchinsky (2015) on 15 adult females taken from four samples containing numerous specimens. In undeformed individuals, the relationship between head length and length of the remaining body parts (except caudal spine) constitutes about 1:1.4. However, because of frequent body deformation in the course of preservation, the body of *Bythotrephes* individuals*,* especially the abdomen, appeared to be constricted or elongated unnaturally, whereas the head was less deformed. Hence, if the size of the post-head body part was smaller or larger than predicted according to this proportion, it was additionally corrected based on head length. In total, eight measurements were calculated and analyzed by application of statistical parameters (M, SD, CV) (Table 2). Similar measurements of males and juvenile specimens were performed (the term "juvenile" is conditionally used here for specimens with only a single pair of postabdominal claws which in fact may be already ovigerous (see Zaffagnini (1964)). The investigation of variability of number and arrangement of setae of tl I–tl IV was made on some specimens from different populations. The setae of thoracic limbs are situated in two parallel rows along their inner side. The setae of the rows closer to the head are termed "anterior", whereas the other ones, closer to postabdomen, are termed "posterior". The number of endopodital segments of a limb was counted, beginning from the proximal one.

In highly morphologically variable Canadian populations, specimens of three distinguishable morphotypes were encountered (Table 3): 1) those with a well-developed bend of the caudal process, bearing two groups of denticles, 2) those with a weakly developed bend of the caudal process, bearing either one small group of denticles or only solitary denticles, and 3) those with either a straight or weakly curved caudal process without denticles (in both these cases, we considered the bend to be absent).

Abbreviations. Morphology: ad = adult parthenogenetic females; BL = body length; CPCL = length of proxi-

mal claws of caudal process; CPL = length of caudal process; gam = gamogenetic females; DS = length of distal endopodital segment of tl I; ICD = distance between postabdominal claws and proximal claws of caudal process (interclaw distance); ICTh = thickness of caudal process between postabdominal claws and proximal claws of caudal process (interclaw thickness); juv = juvenile females or males; PCL = length of postabdominal claws; PS = length of proximal endopodital segment of tl I; TIL = length of the thoracic limb of first pair (tl I); tl I…tl IV = thoracic limbs of first…fourth pairs.

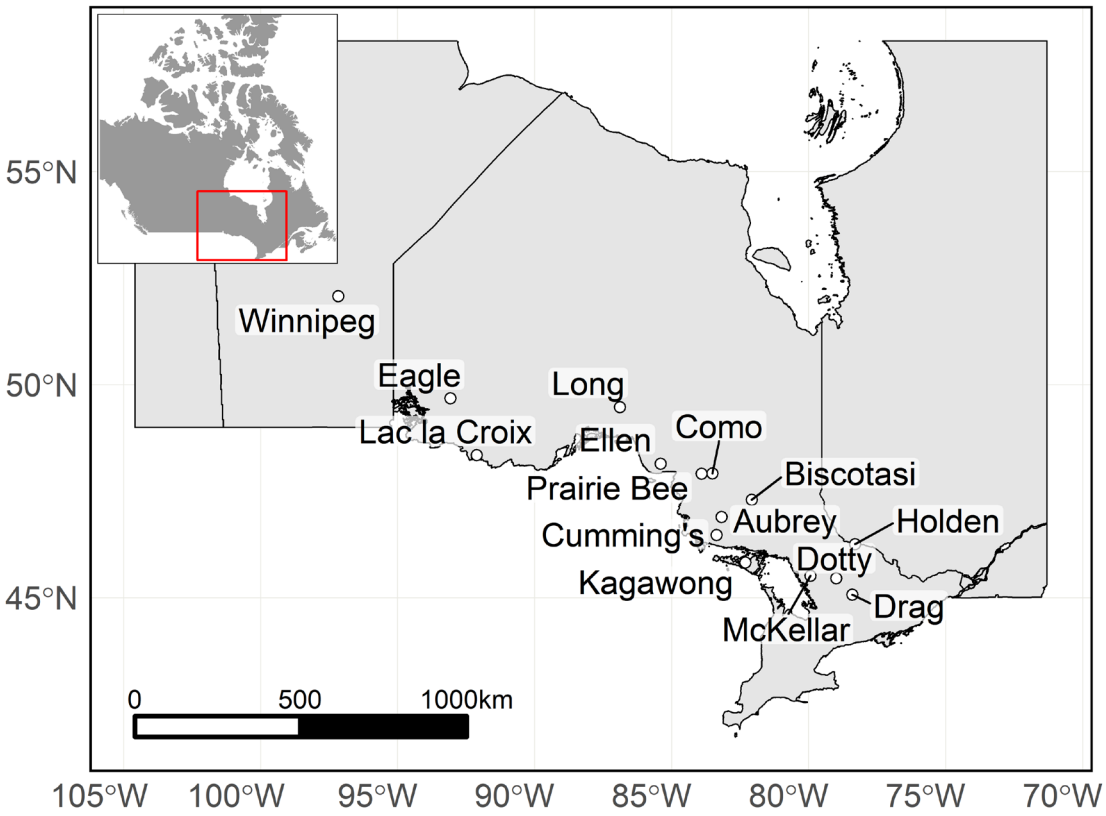

FIGURE 1. Map of the area of the sampled lakes.

TABLE 1. Lake characteristics, based on Ontario Ministry of Natural Resources and Forestry Broad Scale Monitoring Program (Sandstrom *et al*. 2015)

Lake name	Surface area (ha)	Maximum depth (m)	Secchi depth (m)	Total Phosphorus (µg/L)	Conductivity (µS/cm)
Aubrey	2008	23	2.8	8.5	62.6
Biscotasi	4110	24	2.5	9	31.2
Como	1605	26	4.5	8.7	61.5
Cummings	523	39	4.8	3.6	28
Dotty	159	27.1	2	6.2	19.2
Drag	1003	55.5	2.8	5.2	56.5
Eagle	31706	32.8	3.6	8.9	40
Ellen	98	57.6	2.3	5.8	25.8
Holden	7710	99	2	12.8	46.3
Kagawong	5634	34	10.5	5.3	277
Lac la Croix	12209	51	3	9.9	41.8
Long	13499	186.1	4.1	14.3	152
McKellar	88	9.8	3	12.4	44.8
Prairie Bee	1136	24	2.6	4.9	49.8
Winnipeg*	2375000	36	1.4	44	313

* Environment Canada and Manitoba Water Stewardship, 2011. State of Lake Winnipeg: 1999-2007 - Highlights. Published by Environment Canada, Ottawa, Ontario.

Results

Taxonomy

Class Branchiopoda Latreille, 1817

Superorder Cladocera Latreille, 1829

Order Onychopoda Sars, 1865

Family Cercopagididae Mordukhai-Boltovskoi, 1966, emend. Martin et Cash-Clark, 1995

Genus *Bythotrephes* Leydig, 1860

Bythotrephes cederströmii Schödler, 1877

(Figs. 2A–4J)

Here we present only a brief species description. For synonymy and expanded description of the Eurasian representatives of the species see Korovchinsky (2015, 2018) and for detailed description of North American representatives see Martin and Cash-Clark (1995).

Data on body and body parts measurements of specimens from four Canadian populations are presented in Table 2.

TABLE 2. Data on body and body parts measurements of the representatives (adult females) of *Bythotrephes cederströmii* from different Canadian populations (in columns, from top to bottom: range, M, SD, CV)

BL, mm	TIL : BL, %	DS : PS, %	CPL : BL, %	PCL : BL, %	CPCL : BL, %	ICD : BL, %	ICTh : BL, %
1. Lake Prairie Bee, 1.6.2016, n = 15							
2.61–3.30	76.2–87.0	66.7–95.9	213–298	16.9–25.1	14.4–20.0	38.2–49.9	3.2–5.7
3.08	81.7	84.0	259	21.7	16.8	45.9	4.9
	3.3	5.2	19.5	2.3	2.8	4.6	0.7
	4.1	6.2	7.5	10.5	16.6	21.3	13.9
2. Lake Eagle, 9.7.2016, n = 15							
2.62–3.30	70.8–86.1	72.9–100.0	218–286	18.6–29.9	15.2–27.5	28.4–40.9	2.8–5.9
2.91	76.1	82.8	257	24.9	23.0	33.8	5.2
	4.3	7.9	19.1	3.5	3.2	4.4	0.4
	5.6	9.6	7.4	14.0	18.0	13.0	8.2
3. Lake Ellen, 5.8.2017, n = 15							
2.30–3.26	72.4–86.8	78.0–102.5	208–252	14.1–23.7	13.3–21.2	21.1–30.2	4.7–6.9
2.75	78.6	85.7	230	19.2	16.9	24.9	5.8
	3.7	6.6	15.0	2.7	2.5	2.7	0.7
	4.8	7.7	6.5	14.1	14.8	10.8	11.4
4. Lake Winnipeg, 9.8.2018 and 11.8.2018, n = 15							
2.43–4.03	75.0–98.2	78.9–107.4	196–301	18.3–32.2	11.0–28.2	17.0–45.6	4.8–7.7
3.45	83.3	92.8	241	22.0	17.6	31.2	6.3
	6.8	6.7	25.3	3.8	4.2	7.6	1.0
	8.2	7.2	10.5	17.2	23.9	24.4	16.1

Material examined. Canada, Ontario: 1) Lake Prairie Bee (47° 91.2 N; 83° 90.5 W), 1.6.2016, 42 ad, 1 male; 2) Lake Eagle (49° 68.1 N; 93° 06.9 W), 8.7.2016, 8 ad and 9.7.2016, 28 ad, 4 gam, 3 males, 14 juv; 3) Lake Como (47° 91.6 N; 83° 35.7 W), 9.7.2016, 6 ad; 4) Lake Holden (46° 25.4 N; 78° 30.4 W), 31.8.2016, 12 ad, 2 gam, 3 juv; 5) Lake McKellar (45° 51.1 N; 79° 93.1 W), 2.9.2016, 4 ad; 6) Lake Aubrey (46° 89.3 N; 83° 17.9 W), 23.6.2017,

2 ad; 7) Lake Cummings (46° 47.2 N; 83° 35.7 N), 25.6.2017, 11 ad, 1 gam, 1 juv; 8) Lake Biscotasi (47° 30.0 N; 82° 08.2 W), 25.6 and 26.6.2017, 29 ad, 40 juv; 9) Lake Drag (45° 06.9 N; 78° 40.6 W), 28.6.2017, 21 ad, 3 juv; 10) Lake Kagawong (45° 82.9 N; 82° 30.5 W), 6.7.2017, 6 ad, 2 gam; 11) Lac La Croix (48° 34.3 N; 92° 11.2 W), 20.7.2017, 9 ad, 2 juv; 12) Lake Ellen (48° 14.6 N 85° 39.8 W), 5.8.2017, 27 ad, 6 males, 7 juv; 13) Lake Long (49° 47.5 N; 86° 89.1 W), 15.8.2017, 7 ad, 2 juv; 14) Lake Dotty (45° 45.2 N; 78° 99.2 W), 11.9.2017, 5 ad, 3 juv. **Manitoba**: 15) Lake Winnipeg, North Basin (53° 15 N; 97° 53 W), 9.8, 10.8, 11.8, and 14.8.2018, 84 ad, 21 gam, 43 males, 21 juv; 15.8.2018, 32 ad, 1 gam, 9 males, 13 juv.

Description. Female. *General body shape* is typical for the genus (Fig. 2A). Postabdomen bears ventrally a pair of massive and long claws curved apically forward (Fig. 2A, 2F: arrowed). Caudal process is very long and thin, bearing similar claws (Fig. 2F: arrowed) and normally a conspicuous denticulated bend posteriorly (in some populations, it may be either strongly reduced or absent) (Figs. 2F, 2G, 2H, 2I). Body length of females (without caudal process) may reach 4.0 mm or slightly more (in the examined specimens it ranged from 2.30 to 4.03 mm), while the length of the caudal process exceeds the body length considerably.

Head comparatively very large and divided into two parts: a rounded anterior part mostly filled with a large compound eye and a posterior part, bearing dorsally a large saddle-shaped neck organ, swimming antennae and mouth parts. Antennules small and situated on the ventral side of the anterior head part beneath the eye (Fig. 2A).

Swimming antennae comparatively long, with elongated cylindrical basipodite. Of two antennal branches, the lower three-segmented one (endopodite), is slightly longer than upper branch (exopodite). The upper branch is four-segmented and lower branch is three-segmented. Small proximal-most segment of upper branch lacks setae, while other segments possess a row of two-segmented swimming setae of more or less similar size, except the distalmost ones which are shorter. General formula of the antennal setae: 0–1–2–5/1–1–5.

Mouth parts are represented by upper lip (labrum), mandibles, and maxillules (maxilla I). Mandibles are bi-lobed and adapted for biting, with a toothed, blade-like posterior lobe and small anterior lobe (mandibular process) (see Korovchinsky 2018: Figs. 9B, 9C, 9D). Maxillules (mx I) look like two cylindrical structure situated posterior to mandibles, maxillae (mx II) are absent.

Carapace looks like a bag-like structure, strongly modified into closed brood pouch (Fig. 2A) widely connected in its base with dorsal side of thorax. It may be often well developed and massive, being filled by large embryos.

Thoracic limbs of first pair (tl I) are especially long and strong (70.8–98.2 % of body length) (Figs. 2A, 2B). Terminally, the inner side of their protopodite bears a small triangular lobe, known as pseudognathobasic process (see the explanation of the term in Korovchinsky (2015)). The external part of protopodite is longer than internal one and bears apically a small conical outgrowth (Fig. 2B). The first segment of endopodite is long and bears 6–8 (usually 6–7) anterior lateral setae. Distally, this segment bears fairly long denticulated anterior seta and longer posterior finely setulated seta. Second segment of endopodite is conspicuously shorter and bears only two apical setae; comparatively short anterior seta and longer posterior one (Fig. 2B, 3B), occasionally the latter may be rather short (Fig. 3J). The terminal, third segment of endopodite is also long but usually somewhat shorter than first proximal segment, however, sometimes it may be either of the same size or slightly longer (66.7–107.4 % of the latter) and bears apically four long roughly spinulated setae, two of them sit terminally and two subterminally. The limbs of second pair (tl II) are considerably shorter, being their protopodite, again externally, conspicuously longer and provided with a conical outgrowth (Fig. 2A). The first, basal segment of their endopodite bears a row of 5–8 (mostly 6–7) rather long anterior lateral setae (their number also can vary in one individual). Also, there are 1–3 posterior lateral seta of the same type on this segment. Internally, this segment bears a stout cylindrical pseudognathobasic process, possessing some apical prominences of different size and one small, thin seta. The distal, third segment of endopodite of the limb bears four setae, two terminal and two subterminal. The limbs of the third pair (tl III) are generally similar to those of the previous ones, differing in the following details: the external outgrowth of their protopodite is conspicuously larger (Fig. 2A) and lateral anterior and posterior setae (if present) of first segment of endopodite are usually fewer (2–6 and 0–3, respectively). The limbs of the fourth pair (tl IV) (Fig. 2E) are considerably reduced; their protopodite bears slightly spinulated seta sited on a short cylindrical base. The only segment of endopodite has two rows of comparatively short spine-like setae: the external row (group) always consists of two setae, and the internal row of 5–7 setae (usually 6–7), which differ in their appearance and armament.

Abdomen (metasome) (Fig. 2A) is often deformed. It is inconspicuously delimited into two segments, short proximal and long distal with a prominent fold more or less in the middle on the dorsal side. "Postabdomen" consisting of two parts: the last small abdominal segment and the postabdomen *per se* (see Korovchinsky (2015)), which

is comparatively small; the anal opening is situated between postabdominal claws which flare outwards from the midline (Figs. 2A, 2F, 2G, 2H, 3A, 3G, 3N, 4A, 4B, 4I, 4J). The latter are large and stout, quite varying in their size with apical ends normally curved forward (14.1–32.2 %, on average 19.2–24.9 % of body length).

Caudal process is directly connected with postabdomen and then proceeds as a very long spine-like structure quite variable in its length (196.0–301.0 %, on average 230.0–259.0 % of body length), thus surpassing body length in two-three times. It is comparatively thin, and its thickness is also conspicuously variable (2.8–7.7 % of body length). In its proximal half, it bears one or two pairs of claws similar to those on postabdomen which may be either smaller or larger (anterior pair 11.0–28.2 % of body length) (Figs. 2A, 2F, 2G, 2H, 3A, 3G, 3N, 4A, 4B, 4I, 4J). All pairs of claws usually sit quite distantly one from another, however, rarely they may be rather densely situated (Fig. 3G, 4J) (17.0–49.9 %, on average 24.9–45.9 % of body length). In its middle part or more proximally (in 36.7–55.8 % of length of caudal process), the caudal process usually bears a prominent bend (Figs. 2I, 3H, 4D) covered by numerous small curved denticles directed forward, which sit on its dorsal and ventral sides. In specimens from different populations, and even in specimens of one population, this bend can be differently pronounced (Table 3) being either strongly or moderately developed (Figs. 2I, 3H, 4D) or underdeveloped (Fig. 3D) and sometimes absent completely (Figs. 3B, 3C, 3F, 3K, 3L, 4E, 4F). In the latter case, the position of the bend is marked only by a few or even solitary large denticles. In some individuals, the weekly developed bend either lacks denticles (Figs. 3E, 3M, 4G) or can be absent at all, thus the caudal process has appeared to be straight, covered by numerous minute spinulae only (in two latter situations, the bend is considered "absent" in Table 3). Borders separating old molted integuments of caudal process with claws normally are quite conspicuous.

"Juvenile" females. Studied juvenile females did not significantly differ from adults in such parameters as comparative length of caudal process, claws, and tl I. Their postabdominal claws might be curved apically either back (Fig. 4H) or forward.

Gamogenetic females differ from parthenogenetic ones only in presence of large yellow-brownish resting eggs (0.32–0.46 mm in diameter) in their brood pouches (according to Jarnagin *et al*. (2000): Fig. 2—0.41–0.45 mm).

Males on average are smaller than females. Thoracic limbs of first pair (tl I) are of moderate size (60.3–66.4 % of body length) as well as each endopodital segment of them, especially distal one (65.8–67.6 % of proximal endopodital segment), which is slightly swollen proximally (Fig. 2C) and bears on its inner side a small strongly chitinized hook with two inner denticles; a circular area of tiny denticles with softer prominent cuticle in its center is situated under it (Fig. 2D). In adult specimens, the copulatory appendages are small and armed with numerous minute spinulae terminally (Fig. 2E).

Size. Females 2.30–4.03 mm; juvenile females 1.92–2.96 mm; males 2.38–2.92 mm.

Intra- and interpopulation variability. The body size of specimens within and between populations varied in some degree, the largest of them were observed in Lake Winnipeg (Table 2). The specimens of Lake Winnipeg had on average the longest tl I although the differences in this parameter between populations were insignificant. The same is true for the length of distal endopodital segment. The comparative length of caudal process of individuals of all studied populations also seems rather similar being, on average, slightly shorter in Lake Ellen. The individuals from Lake Ellen also had shorter postabdominal claws whereas those from Lake Eagle had longer claws of caudal process. Interclaw distance was largest in the representatives from Lake Prairie Bee which probably is directly connected with their smallest interclaw thickness. The latter parameter was largest in Lake Winnipeg. In general, the highest variability (CV) was observed in postabdominal and caudal claws, interclaw distance, and interclaw thickness.

The bend of the caudal process was well developed in all studied specimens from lakes Eagle and Holden, and in most of them from lakes Drag (92 %) and Winnipeg (95 %) (Table 3). On the other hand, it was absent in most specimens of lakes Prairie Bee, Ellen, and Biscotasi where a weakly developed bend was observed in the smallest number of individuals (3–23 %).

In most individuals of all populations, the apical end of claws, especially those of caudal process, were curved forward although in few of them it might be straight (Figs. 3A, 3G, 4I, 4J). No individuals were found with the claws of caudal process turned upwards.

Remarks. A quite detailed description of the North American representatives of the species was provided previously by Martin and Cash-Clark (1995). It fully coincides with the above description and well supplements it with respect to SEM micrographs of some structures.

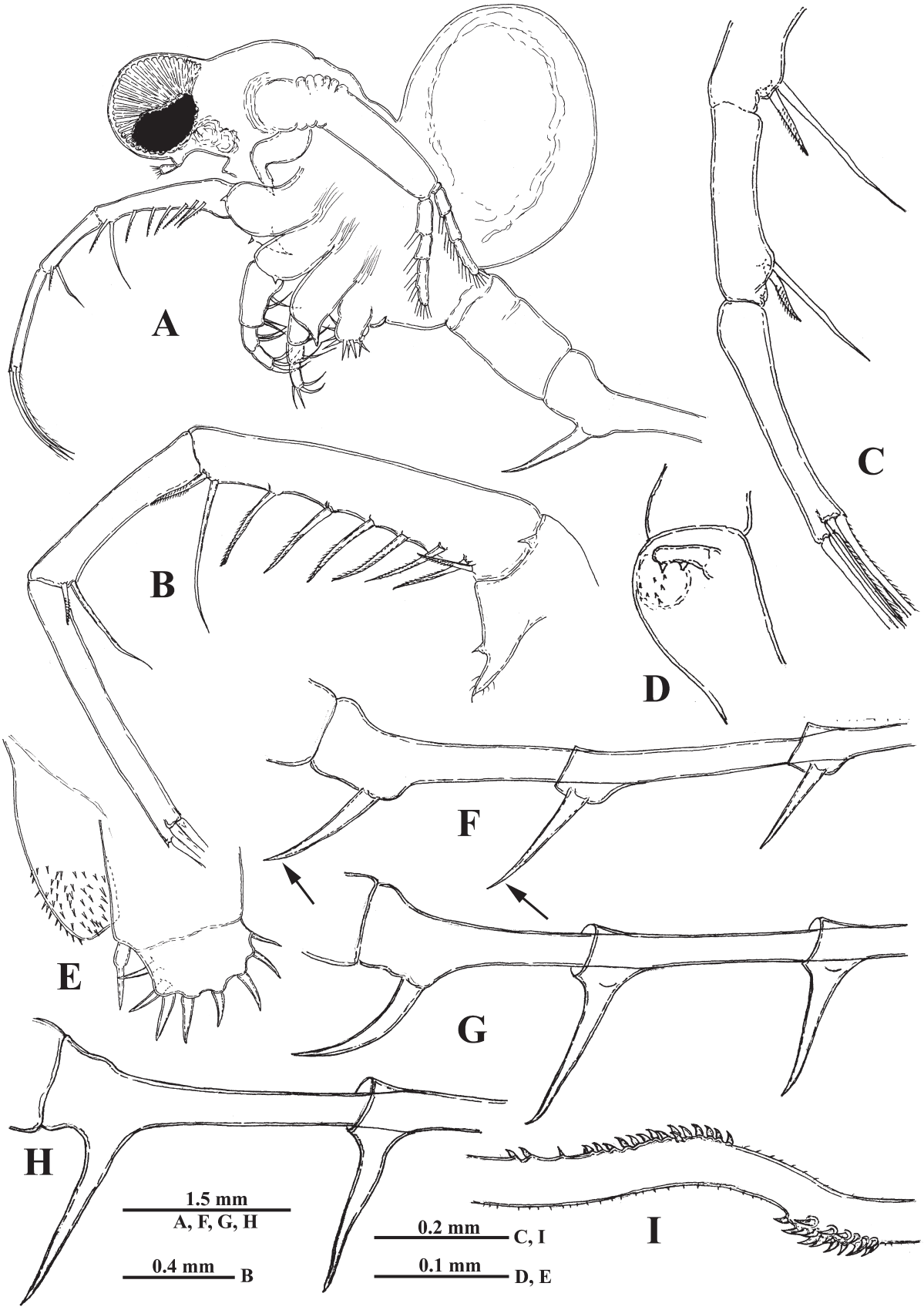

FIGURE 2. *Bythotrephes cederströmii,* females and males (Canada: A—Lake Eagle; B—Lac La Croix; C–I—Lake Prairie Bee). Female: A, general lateral view. B, thoracic limb of first pair (tl I). F–H—claws of postabdomen and caudal process. I—bend of caudal process. Male: C, thoracic limb of first pair (tl I). D, inner proximal part of third segment of tl I with clasping hook. E, thoracic limb of forth pair (tl IV) and copulatory appendage.

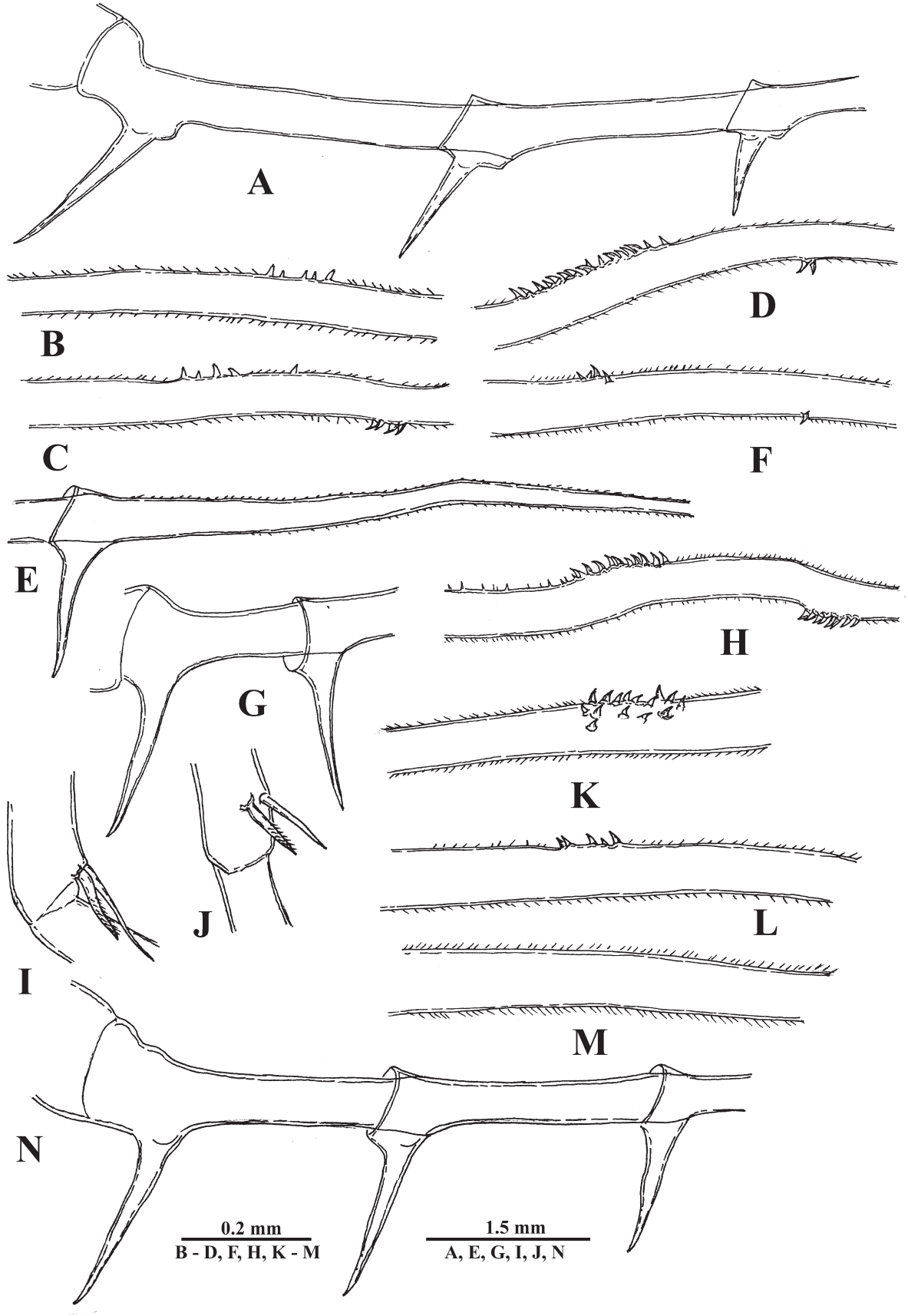

FIGURE 3. *Bythotrephes cederströmii,* females (Canada: A–D—Lake Prairie Bee; E–G—Lake Ellen; H—Lake Long; I–K—Lake Dotty; L, M—Lake Biscotasi; N—Lake Cummings). A, G, N, claws of postabdomen and caudal process. I, proximal part of caudal process. B–F, H, K–M, bend of caudal process. I, J, distal setae of second endopodital segment of thoracic limbs of first pair (tl I).

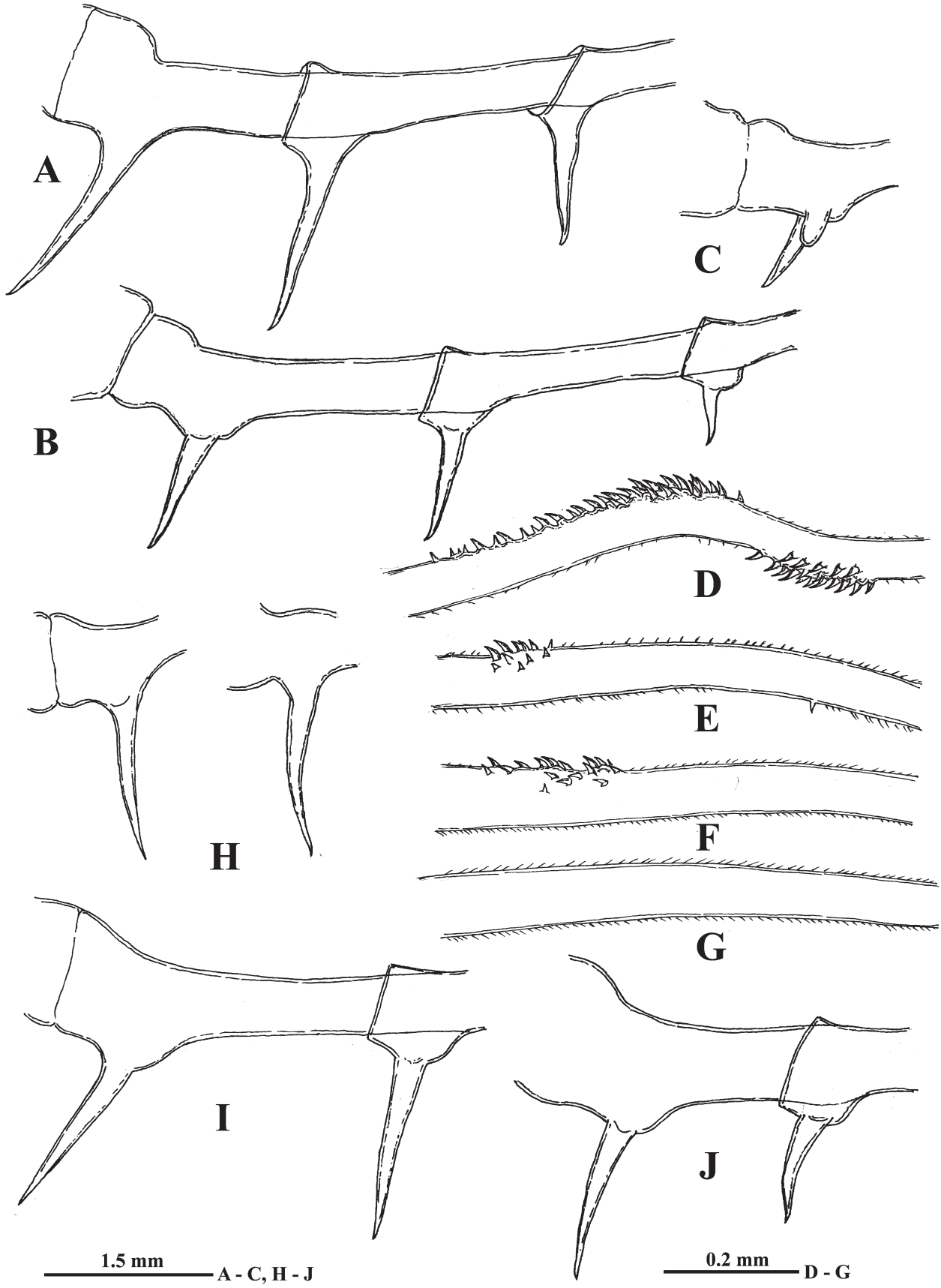

FIGURE 4. *Bythotrephes cederströmii,* females (Canada: A—Lake Aubrey; B, C—Lake Kagawong; D—Lac La Croix; E–J—Lake Winnipeg). A, B, I, J, claws of postabdomen and caudal process. C, damaged postabdominal claw. H, postabdominal claws of juvenile females. D–G—bend of caudal process.

In general, the variability of the North American *Bythotrephes* seems high, especially that of caudal process and claws, but nevertheless it definitely fits within that of *B. cederströmii* (see Korovchinsky 2015, 2018) (for more detailed comparison of the American and Eurasian representatives of the species, see further). Despite the fact that the presence of large, massive postabdominal and caudal process' claws apically curved forward are especially characteristic of this species, the presence of a small number of specimens with apically straight claws also falls within the range of species variability (see Korovchinsky 2018: Fig. 14A).

Differential diagnoses. *B. cederstromii* differs from all other species of the genus firstly in the presence of large, massive (rarely comparatively small) postabdominal claws and claws of caudal process, flared outwards, and usually curved forward apically. In specimens from different American populations, and even in specimens of one population, the bend of caudal process can be differently pronounced being either strongly or moderately developed, or underdeveloped and sometimes absent completely (see Table 3), wherewith the caudal process looks straight. Normally, the caudal bend is supplied with two groups of small denticles, otherwise the position of the bend may be marked either only by a few or even solitary large denticles or they lack at all.

B. longimanus s.str. differs from *B. cederstromii* in presence of usually longer tl I with longer third distal endopodital segment. The anterior distal seta of proximal segment of endopodite and both apical setae of the following second segment of endopodite of tl I are small, reduced up to minute thorns or sometimes may be absent completely. Adults always have only two pairs of relatively large (~5.0–13.0 % of body length) and straight claws directed downwards or/and slightly backwards, and situated close one to another (Korovchinsky (2015)).

TABLE 3. Percentage (%) of specimens of *Bythotrephes cederströmii* from some populations possessing variously developed bend of caudal process

Bend of caudal process:	Lakes (number of specimens)						
	Prairie Bee (43)	Eagle (57)	Holden (16)	Ellen (38)	Biscotasi (69)	Drag (24)	Winnipeg (216)
well developed	14	100	100	8	23	92	95
weakly developed	23	-	-	21	10	8	3
absent	63	-	-	71	67	-	2

Discussion

Thus, the invasive species of *Bythotrephes* in North America, analyzed in the aforementioned Canadian lakes, appears to be *B. cederströmii* s. str., as originally, though tentatively and uncertainly, identified (see Bur *et al*. 1986; Lange & Cap 1986; Evans 1988; Litvinchuk 2007, etc.). Thus, the recent name used for the North American *Bythotrephes*, *B. longimanus,* must be rejected.

Judging by figures and photographs published in some papers on North American representatives (Evans 1988; Berg & Garton 1988; Keilty 1988, 1990; Barnhisel 1991a, b; Yurista 1992; Burkhardt 1994; Martin & Cash-Clark 1995; Yan *et al*. 2011; Korosi & Smol 2012), the same and only species, *B. cederströmii,* was recorded in all Great Laurentian Lakes and a few Canadian lakes. Probably a similar conclusion is correct for all populations of *Bythotrephes* occurring in North American inland waters.

Earlier it was hypothesized that both *B. cederströmii* and interspecific hybrids *B. brevimanus* x *B. cederströmii* could invade the North American inland waters (Litvinchuk 2002; Korovchinsky 2015, 2018). Based on recent morphological investigations of the latter (Korovchinsky 2019) this suggestion must be ruled out. No sample of *Bythotrephes* studied from Canada contained the individuals of characteristic hybrid form appearance. Theoretically speaking, the formation of hybrids in North American waters is impossible simply due to the absence of the second parental species, *B. brevimanus* Lilljeborg, 1901. The latter one did not yet colonize the North American continent, although it inhabits the same native region (Eastern Baltic region) from where the invasive specimens of *B. cederströmii* were probably introduced. Possibly the establishment of *B. brevimanus* in North America has been prevented by the founder effect of its abundant congener that widely colonized the region first. The same may be true with regard to the colonization of North America by hybrids *B. brevimanus* x *B. cederströmii.* Moreover, these hybrids can possess the weakened sexual reproduction and consequently the comparatively low dispersal ability and survival probability during harsh environmental conditions as it is known in hybrid forms of *Daphnia* (see Spaak *et al*. 2004; Keller *et al*. 2007).

The Eurasian representatives of *B. cederströmii* are morphologically variable, especially in the structure of their caudal process and claws (Korovchinsky 2018). The considerable morphological variability in studied individuals was also noted by some North American researchers (Evans 1988; Sprules *et al*. 1990; Burkhardt 1994; Bilkovic & Lehman 1997). This is likely what caused the uncertainty of species identification, especially because a comprehensive morphological investigation based on usage of abundant European material was lacking. This obstacle has been removed owning to available data of the recent revisions (Korovchinsky 2015, 2016, 2018, 2019) describing the taxonomic richness of the genus *Bythotrephes* and the geographic distribution of particular species.

The comparison of Canadian and Eurasian representatives of *B. cederströmii* has revealed that the former have larger body size, longer thoracic limbs of the first pair (tl I), shorter caudal spine and postabdominal claws, and smaller interclaw distance. Their morphological parameters generally are either less variable or have the same level as those of the Eurasian individuals. This coincides with data suggesting comparatively less genetic variability of North American bythotrepheses (see Weider 1991; Berg & Garton 1994). On the other hand, the expression of the bend of the caudal process seems to be more variable in Canadian *B. cederströmii,* than in Eurasian specimens: in some Canadian populations, most of individuals (63–71 %) lacked the band at all (Table 3) which was not the case in the Eurasian ones. However, more material from more populations is needed for the proper intercontinental comparisons.

B. cederströmii have proved to be the most widely distributed species of the genus. In Eurasia it is distributed from Ireland and Scandinavia to Central Yakutia in Eastern Siberia while in North America it occupies a large number of lakes over the huge territory of eastern parts of the continent (Korovchinsky 2018; Kerfoot *et al*. 2016; Jansen *et al*. 2017). Thus, it may be considered a species having almost Holarctic distribution.

In North America, *B. cederströmii* is rightly considered "the world's best studied invasive zooplankter" (Bollens *et al*. 2002; Yan *et al*., 2011) which is especially true regarding morphology (Martin & Cash-Clark 1995; Korovchinky 2015, 2018) and ecology of the local representatives of the species. At the same time, the Eurasian populations of the species remain ecologically poorly studied. And of course, the molecular-genetic studies of the species all over its range are highly desirable.

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