

Alloglossidium demshini sp. nov. (Digenea: Macroderoididae) from leeches in Minnesota

Vasyl V. Tkach*, Stephen E. Greiman and Kayla R. Steffes

Department of Biology, University of North Dakota, Grand Forks, North Dakota 58202, U.S.A.

Abstract

Alloglossidium demshini sp. nov. is described based on specimens from leeches *Haemopsis grandis* collected in northwestern Minnesota. The new species is morphologically closest to *Alloglossidium schmidtii*. The two species can be readily differentiated based on several morphological characters. The cirrus sac in *A. schmidtii* is almost entirely situated anterior to the ventral sucker while in *A. demshini* sp. nov. it is situated dorsal to the ventral sucker and its proximal end almost reaches the posterior end of ventral sucker or extends posterior to it. The new species has a prepharynx that is substantially longer than the esophagus while in *A. schmidtii* the situation is the opposite. The two species also differ in the position of the ovary and the position of the testes and vitelline fields in relation to the ends of the ceca. *Hirudineatrema oschmarini* described from leeches in Eastern Palaearctic and *Alloglossidium richardsoni* described in North America demonstrate great morphological similarity. Nevertheless, *Hirudineatrema* cannot be synonymized with *Alloglossidium* at this point because of several peculiar morphological features of *H. oschmarini* such as a V-shaped excretory bladder and the apparent presence of a true seminal receptacle in the latter species. These features need to be re-evaluated before any taxonomic decision can be made.

Keywords

Digenea, *Alloglossidium*, *Alloglossidium demshini* sp. nov., *Haemopsis grandis*, leeches, Minnesota.

Introduction

Alloglossidium Simer, 1929 is a genus of plagiorchoid digeneans parasitic in fishes, leeches, and crustaceans in North America and the Eastern Palaearctic (Font 1980, Brooks 2003, Font and Lotz 2008, Tkach and Mills 2011). While some *Alloglossidium* are intestinal parasites of fish, other species mature in the antennal glands or body cavity of crustaceans and the intestinal lumen of leeches. Schmidt and Chaloupka (1969) described what they thought was the first member of the genus or any digenean living and maturing in the intestine of leeches. They clearly were not aware of the paper of Demshin (1968) who was in fact the first to discover adult digeneans *Hirudineatrema oschmarini* Demshin, 1968 from the intestine of leeches *Haemopsis sanguisuga* (Linnaeus, 1758) in the Russian Far East. In Schmidt and Chaloupka's defense, Demshin's paper was published in a non-periodical edition that would be very difficult to get in the middle of the Cold War. Brooks (2003) in his insightful paper/essay "Lessons from a quiet classic" has described in detail the history of the discovery of various species of *Alloglossidium* with a particular emphasis

on the species capable of attaining maturity inside invertebrate hosts. Although Demshin's paper of 1968 was cited by Brooks (2003), Demshin was not given the credit as the first discoverer of adult digeneans in leeches. Demshin has also published a substantial amount of other research on parasites of annelids and other animals (e.g., Demshin 1965, Oshmarin *et al.* 1970, Demshin and Leonov 1975, Demshin 1976, Demshin 1985) and certainly deserves his rightful place in the ranks of quiet classics who developed and advanced our knowledge of this unique digenean group and other parasites.

There was a steady flow of descriptions of new *Alloglossidium* species from the early 1970s through the mid-1990s. Some of these species were initially placed in other genera such as *Hirudicolotrema* Fish and Vande Vusse, 1976 and *Alloglossoides* Corkum and Turner, 1977 which were synonymized with *Alloglossidium* (Smythe and Font 2001, Brooks 2003). However, after the description of *Alloglossidium greeri* from crayfish *Cambarellus shufeldtii* by Font (1994) it took another 17 years until Tkach and Mills (2011) described the next species *Alloglossidium fontii* from bullhead catfishes in Minnesota. These authors have also used a combination of molecular and mor-

*Corresponding author: vasyi.tkach@email.und.edu

phological data to differentiate the new species and resurrect the type species of the genus, *Alloglossidium kenti* Simer, 1929.

In the course of parasitological study of leeches in north-western Minnesota we have collected numerous specimens of digeneans belonging to 5 different *Alloglossidium* species. One of the species collected from *Haemopsis grandis* Verrill, 1874, demonstrated clear morphological differences from all *Alloglossidium* previously known in leeches or other hosts. The discovery was surprising considering the history of rather intensive investigations of *Alloglossidium* in leeches in the upper Midwest and neighboring region in Canada (Timmers 1979, Vande Vusse 1980, Neumann and Vande Vusse 1976, Brooks 2003). In this work we describe the new species and provide its differentiating diagnosis from congeners.

Materials and Methods

Two leech species, *Haemopsis grandis* and *Haemopsis mar-morata* (Say, 1824) were collected in Nelson Slough impoundment, East Park Wildlife Management Area, Marshall Co., Minnesota (48°29'35.34"N 96°20'54.39"W) and examined for parasites. Identification keys by Klemm (1982) were used for leech identification. Digeneans belonging to the new species were found only in *H. grandis*.

Live worms were rinsed in saline, briefly examined prior to fixation, killed with hot water, and fixed in 70% ethanol. Some specimens were fixed in 95% ethanol for molecular analysis. We do not provide further details of molecular work here because we do not have DNA from the morphologically closest species *Alloglossidium schmidtii* Timmers, 1979. In the absence of sequences from the latter species the sequence differentiation and comparison would not be informative enough. Specimens used for morphological study were stained with aqueous alum carmine or Mayer's hematoxylin, dehydrated in a graded ethanol series, cleared in clove oil (after carmine staining) or methyl salicylate (after hematoxylin staining), and mounted permanently in Damar gum. Measurements were taken from a DIC-equipped Olympus BX-51 microscope using Rincon software (v. 7.1.2, Imaging Planet, Goleta, California). Drawings were made with the aid of a drawing tube on a Leica DM5000 compound microscope. All measurements are in micrometers.

The type specimens of the new species were submitted to the U.S. National Parasite Collection (Beltsville, Maryland) under accession numbers USNPC 106813-106814.

The specimens for scanning electron microscopy (SEM) were fixed in 70% ethanol, dehydrated in a graded series of ethanol, and dried with hexamethyldisilazane (Ted Pella Inc., Redding, California) as a transition fluid. The specimen was mounted on an aluminum stub using conductive double-sided tape, coated with gold-palladium, and examined with the use of a Hitachi 4700 scanning electron microscope (Hitachi U.S.A., Mountain View, California) at an accelerating voltage of 5–10 kV.

Results

Alloglossidium demshini sp. nov. (Figs 1–3)

Description based on 19 adult worms (one of them incomplete because its posterior end was used for DNA extract). Measurements of the holotype are given in the text, while measurements of the entire type series are given in Table I. Body elongate, 2618 long $\times$ 501 wide; body width measured at level of ventral sucker where it is usually greatest. Body width to length ratio 1:5.2. Tegument at anterior end of body is armed with small spines; tegumental spines were not observed posterior to level of pharynx under light microscope, although under SEM (Fig. 3E, F) they are seen to reach level of ventral sucker. Oral sucker 157 $\times$ 171, subterminal, similar in size to ventral sucker. Ventral sucker 173 $\times$ 185, situated at approximately 1/5 of body length from anterior end; distance from center of ventral sucker to anterior body end 528. Prepharynx short, 103, usually dilated at its posterior end, somewhat longer than esophagus. Pharynx muscular, 106 $\times$ 109. Esophagus short, 78, cecal bifurcation near anterior margin of ventral sucker, 438 from anterior end of body. Ceca terminate just posterior to middle of body, 1070 from posterior body end.

Testes 2, small, oval, intercecal in middle of body, tandem or slightly diagonal; anterior testis 156 $\times$ 85; posterior testis 166 $\times$ 94; distance between testes 1 testis size or less. Posterior margin of posterior testis at level of ends of ceca, may be slightly anterior or posterior to it. Cirrus-sac, 236 $\times$ 73, mostly overlapped by ventral sucker, its base may slightly extend beyond posterior margin of ventral sucker or just above posterior margin of ventral sucker; cirrus sac contains a bipartite seminal vesicle, prostatic duct, and associated prostatic cells near distal end; no evaginated cirrus was observed. Proximal part of internal seminal vesicle longer than distal part. Genital pore immediately anterior to ventral sucker, slightly submedian, sinistral.

Ovary 236 $\times$ 157, much larger than testes, pretesticular, median, longitudinally elongate, situated 138 posterior to ventral sucker, approximately halfway between anterior testis and ventral sucker. Ootype surrounded by Mehlis' gland, situated just ventral and antero-sinistral to ovary. True seminal receptacle is absent. Sperm is stored in proximal portion of uterus. Laurer's canal opens on dorsal side of body at level of posterior margin of ovary. Vitellarium consisting of numerous small vitelline follicles arranged in 2 lateral fields that may merge dorsally at level of ventral sucker and/or at level of posterior margin of ovary. Anterior margins of vitelline fields reach oral sucker or at least the level of anterior margin of pharynx; posterior ends of vitelline fields approximately at the level of ends of ceca, usually slightly anterior to them. Uterus occupying much of postovarian space between vitelline field and most of the hind body posterior to ends of ceca and vitelline fields. Proximal and distal parts of uterus usually overlapping ovary, testes and ventral sucker ventrally. Metraterm well differentiated, with muscular walls, begin-

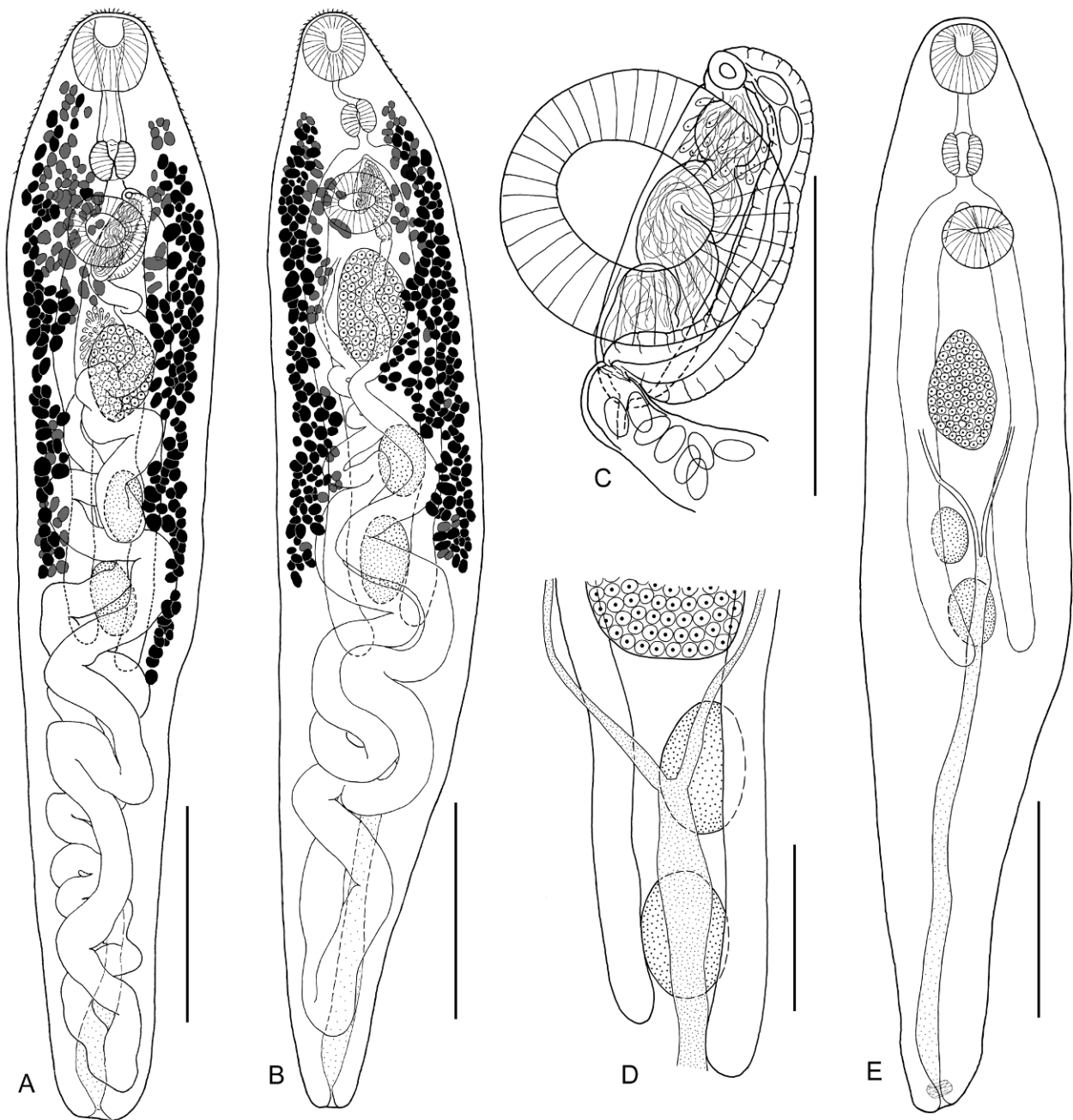


Fig. 1. *Alloglossidium demshini* sp. nov. **A** – ventral view of the holotype. **B** – ventral view of a paratype. **C** – ventral view of ventral sucker region of the holotype showing relative position of cirrus sac, metraterm and ventral sucker. **D** – fragment of a paratype showing relative position of the distal end of excretory bladder, testes, ceca and ovary. **E** – ventral view of a paratype showing shape of the excretory bladder and its position relative to gonads; note left and right common excretory canals beginning at the anterior end of the excretory bladder which makes an impression of a Y-shaped structure when the bladder is not filled. Uterus, cirrus-sac and vitellarium not shown. Scale bars = 500 μ m (A, B, E), 200 μ m (C), 250 μ m (D)

ning at level of posterior end of cirrus sac, 248 in length. Eggs numerous, $26\text{--}31 \times 13\text{--}16$.

Excretory pore terminal; excretory bladder I-shaped, winding between testes; distal end at level of anterior testis; left and right common excretory canals begin from anterior end of excretory bladder.

Taxonomic summary

Type host: *Haemopsis grandis* Verrill, 1874 (Annelida: Clitellata: Haemopidae).

Site of infection: intestine.

Type locality: Nelson Slough impoundment, East Park

Wildlife Management Area, Marshall Co., Minnesota (48°29'35.34"N 96°20'54.39"W).

Type specimens deposited: holotype, USNPC 106813 (labelled: *Haemopsis grandis*, intestine, Marshall County, MN, 19 August 2012, coll. V. Tkach); paratypes USNPC 106814 (18 slides, labelled identically to the holotype).

Etymology: The species is named in honor of Nikolai Demshin who was the first researcher who found and described adult digeneans in the intestine of leeches.

Remarks

Based on the combination of morphological features namely the cuticular spination, tandem testes, pretesticular ovary, well-developed cirrus-sac containing a bipartite internal seminal vesicle, vitelline follicles arranged in two lateral fields, I-shaped excretory bladder reaching the level of ovary, and absence of a seminal receptacle, the new species is fully consistent with the diagnosis of *Alloglossidium*.

Alloglossidium hamrumi Neumann and Vande Vusse, 1976 has very different body proportions compared to the new species. It is much thinner and much more elongated, and has an ovary smaller than the testes (Neumann and Vande Vusse 1976). Besides, unlike the new species, the vitelline fields in *A. hamrumi* reach only the posterior end of the pharynx and the ceca extend further posteriorly terminating at about one quarter the body length from the posterior end of body.

Alloglossidium hirudicola Schmidt and Chaloupka, 1969 differs from *A. demshini* sp. nov. by the ovary being substantially smaller than the testes. The testes in *A. hirudicola* are much larger, spherical rather than elongated; the ventral sucker is distinctly larger than the oral sucker, and the anterior margin of the vitelline fields do not reach as far anteriorly as in the new species (Schmidt and Chaloupka 1969).

Alloglossidium macrobdellensis Beckerdite and Corkum, 1974 can be easily differentiated from the new species by the much more elongated body, much smaller suckers, longer cirrus sac, significantly longer prepharynx, an ovary that is

Table I. Metric data for *Alloglossidium demshini* sp. nov.

Characters	n	Min-Max	Mean	StD	CV*
Body length	18	2112–3704	2726.3	447.7	16.4
Body width	19	142–680	491.6	130.4	26.5
Body W/L ratio	17	1:4.3–1:6.5	1:5.6	0.6	11.2
Ventral sucker to ant. end	19	348–654	506.6	82.5	16.3
Oral sucker length	19	109–207	165.5	25.8	15.6
Oral sucker width	19	85–218	159.2	33.4	21.0
Prepharynx length	19	40–130	79.1	24.6	31.1
Pharynx length	19	85–138	112.4	16.4	14.6
Pharynx width	19	86–137	105.3	15.2	14.4
Esophagus	19	39–102	64.4	20.0	31.0
Cecal bifurcation to ant. end	19	311–566	447.2	76.5	17.1
Ventral sucker length	19	110–230	168.2	31.3	18.6
Ventral sucker width	19	130–257	187.2	32.8	17.5
Cirrus sac length	19	158–310	213.8	40.8	19.1
Cirrus sac width	19	42–100	73.6	13.8	18.7
Genital pore to ant. end	19	294–590	426.3	71.7	16.8
Metraterm	19	168–312	220.1	37.3	16.9
Vitelline field to post. end	18	934–1808	1174.8	227.3	19.3
Ovary length	19	199–365	265.0	47.0	17.7
Ovary width	19	119–242	162.8	28.5	17.5
Ovary to ventral sucker	19	31–203	114.9	43.2	37.6
Anterior testis length	19	114–228	152.2	27.4	18.0
Anterior testis width	19	63–148	101.3	19.8	19.6
Posterior testis length	19	134–245	187.4	29.6	15.8
Posterior testis width	19	71–167	104.5	22.3	21.4
End of ceca to post. end	18	850–1681	1166.7	217.9	18.7
Egg length	47	27–31	28.8	1.1	3.7
Egg width	47	15–18	16.3	0.7	4.2

*Coefficient of variation.

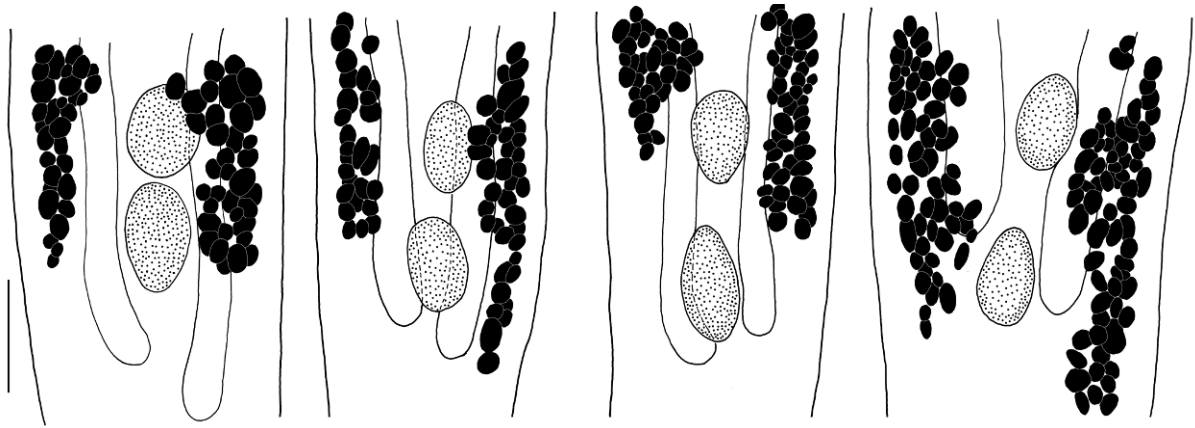


Fig. 2. *Alloglossidium demshini* sp. nov. Variability in the relative position of testes, posterior ends of ceca and posterior margins of vitellaria. Scale bar = 200 μ m

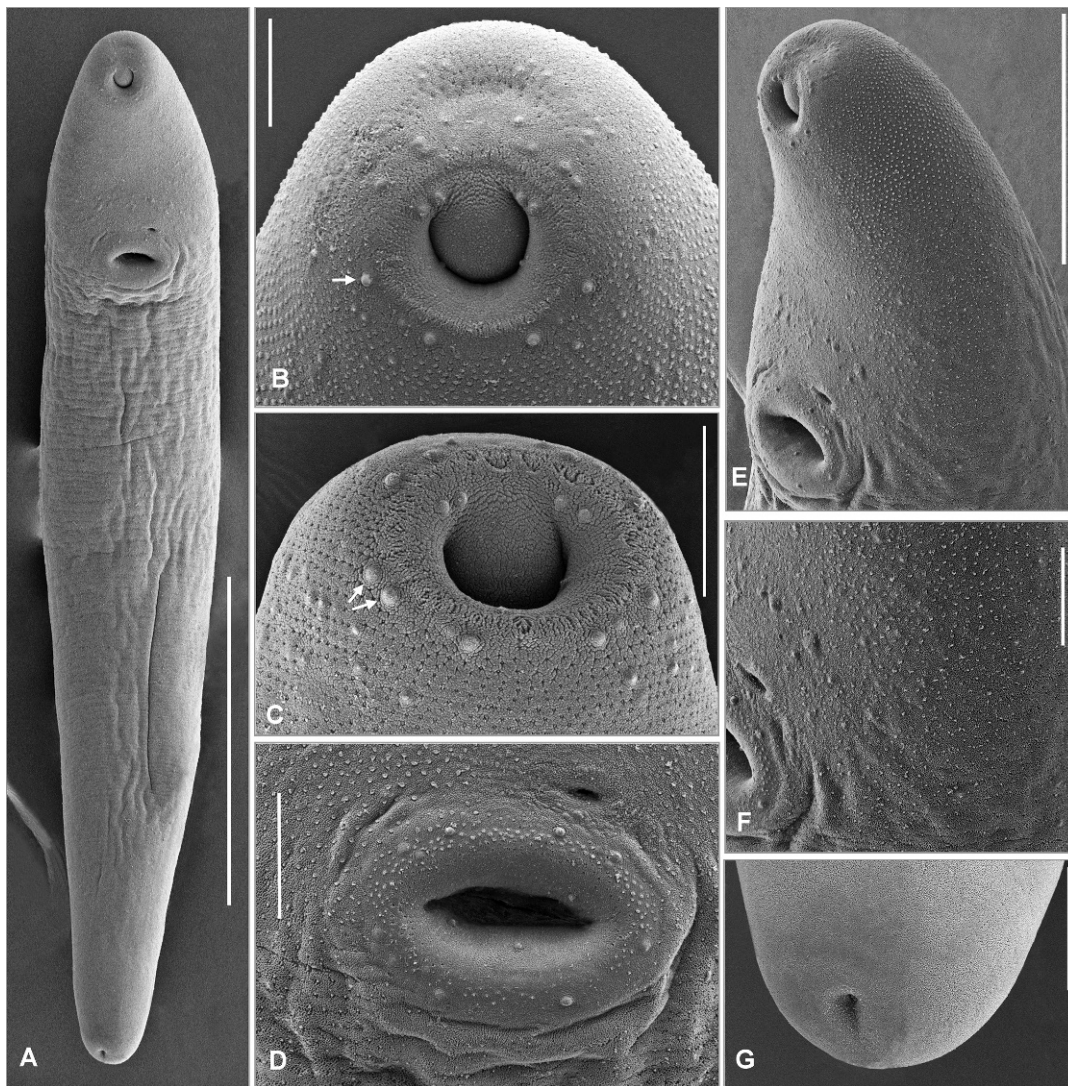


Fig. 3. SEM images of *Alloglossidium demshini* sp. nov. **A** – total view. **B,C** – oral sucker region. Note the inconsistencies in the distribution and number of circumoral sensory papillae (indicated by arrows). **D**. Ventral sucker. **E** – lateral view of forebody. **F** – lateral view of body at level of oral sucker showing minute spines reaching the level of posterior margin of ventral sucker. **G** – posterior end of body with excretory pore. Scale bars = 500 μ m (**A**), 50 μ m (**B–D**, **F**, **G**), 200 μ m (**E**)

smaller than testes and testes situated much more posteriorly. In addition, the vitelline fields in *A. macrobdellensis* only reach the intestinal bifurcation anteriorly and terminate posteriorly well anterior to the end of the ceca and anterior to both testes (Beckerdite and Corkum 1974).

Alloglossidium turnbulli Neumann and Vande Vusse, 1976 substantially differs from *A. demshini* sp. nov. in having a much more elongated body shape, much longer prepharynx, much greater distance between ovary and testes and most importantly, the vitelline fields not reaching the anterior margin of the ventral sucker (Neumann and Vande Vusse 1976).

The new species is morphologically closest to *Alloglossidium schmidtii*. Similarly to *A. demshini* sp. nov., *A. schmidtii* has vitelline follicles reaching the oral sucker, an ovary that is larger than the testes in size, and suckers that are approximately equal in size (Timmers 1979, Vande Vusse 1980). However, the two species can be readily differentiated based on multiple morphological characters. The cirrus sac in *A. schmidtii* is relatively smaller than in the new species and almost entirely situated anterior to the ventral sucker while in *A. demshini* sp. nov. it is situated dorsal to the ventral sucker and its proximal end almost reaches the posterior end of ventral sucker or extends posterior to it. The prepharynx in the new species is substantially longer than the esophagus while in *A. schmidtii* the situation is the opposite. The ovary in *A. schmidtii* is situated immediately postero-dextral to the ventral sucker while in the new species it is median and situated at some distance from the ventral sucker. The most discriminative feature between the two species is the position of the testes and vitelline fields in relation to the ends of the ceca. In the new species the ceca extend only slightly past the posterior testes or do not even reach the posterior margin of the posterior testis while in *A. schmidtii* they extend far posteriorly to the testes. Although the egg sizes in *A. demshini* and *A. schmidtii* somewhat overlap, the average egg size in the new species is somewhat smaller (25–31, average 27 vs 27–33, average 29 in *A. schmidtii*).

Discussion

Tkach and Mills (2011) examined numerous genetically marked specimens of 4 species of *Alloglossidium* parasitic in catfishes. These authors have demonstrated that the anterior extent of the vitelline fields was one of the most reliable differentiating features. Among previously known species of *Alloglossidium* from leeches, only *A. schmidtii* has vitelline fields reaching the oral sucker similar to the condition in the new species, however, the two species have many other distinct distinguishing features.

Despite the high quality of mounted specimens and use of differential interference contrast optics, we were unable to detect tegumental spines in the new species posterior to the level of the pharynx. However, SEM has revealed very small spines reaching the level of the posterior margin of the ventral sucker

in all three specimens examined (Figs 3E, F). Among other morphological features observed under SEM the circumoral sensory papillae (Figs 3B, C) were of interest. The number and arrangement of these papillae are usually stable within species and even within a genus of digeneans. However, we observed variability in both relative position and number of papillae between different specimens of *A. demshini* sp. nov. studied under SEM (Figs 3B, C). Our discovery of the new *Alloglossidium* species from leeches after more than two decades since the description of the previous species suggests that denser geographic sampling may result in new findings even from leech species that were the subject of several previous studies in the same general region. Based on the similarities in several important morphological features between the new species and *A. schmidtii*, especially that the ovary is larger than the testes in size and the vitelline fields reach the oral sucker, we hypothesize that the new species is phylogenetically most closely related to *A. schmidtii*. However, in the absence of DNA from *A. schmidtii* we were unable to test this hypothesis using sequence data. Obtaining DNA sequences from all known species of *Alloglossidium* from fishes and invertebrate hosts is critical for our understanding of the evolutionary history of this extremely interesting digenean group as well as for clarification of existing taxonomic questions within the genus.

Demshin (1968) described the first digenean species maturing in the leech intestine, *Hirudineatrema oschmarini*. He established a new genus and a new subfamily Hirudineatreminae for this species. *Hirudineatrema oschmarini* is morphologically closest to *Alloglossidium richardsoni* (Fish and Vande Vusse, 1976) which was initially placed in its own genus *Hirudicolotrema* (Fish and Vande Vusse 1976) and later synonymized with *Alloglossidium* by Smythe and Font (2001). While it is likely that *H. oschmarini* belongs to *Alloglossidium*, we refrain from synonymization of *Hirudineatrema* with *Alloglossidium* because of several morphological characters of *H. oschmarini* that need confirmation. According to the original description, the excretory vesicle in *H. oschmarini* is V-shaped while in *Alloglossidium* it is I-shaped (e.g., Fig. 1D, E). Although the original description of *H. oschmarini* or the diagnosis of *Hirudineatrema* do not say anything about the seminal receptacle, the drawing shows a regular spherical seminal receptacle while members of *Alloglossidium* lack a true seminal receptacle and possess a uterine seminal receptacle instead. Lastly, Demshin (1968) reported some sort of ancillary glands positioned at the anterior margin of each testis. The nature of these glands remains unknown. Demshin (1968) did not include any information on the deposition of the type material and *H. oschmarini* has not been reported by anyone else after the original description. Therefore, we consider any nomenclature changes regarding this species and genus premature until the type material can be located and re-examined or new material collected. Obtaining molecular data for *H. oschmarini* and inclusion of its sequences into a phylogenetic analysis would provide an additional important source of in-

formation that will help to clarify the systematic position of this very interesting species.

Acknowledgements. The authors are grateful to Alan Nelson and Susana Rios for their assistance in field collecting of leeches and to Mike Kinsella for his useful remarks regarding our manuscript.

References

- Beckerdite F.W., Corkum K.C. 1974. *Alloglossidium macrobdellensis* sp. n. (Trematoda: Macroderoididae) from the leech, *Macrobdella ditetra* Moore, 1953. *Journal of Parasitology*, 60, 434–436. DOI: <http://dx.doi.org/10.2307/3278357>.
- Brooks D.R. 2003. Lessons from a quiet classic. *Journal of Parasitology*, 89, 878–885. DOI: 10.1645/GE-3226DF.
- Demshin N.I. 1965. On biology of *Porrocaecum skrjabinensis* Mosgovoi, 1949. In: (Ed. P.G. Oshmadin) *Parasitic worms of domestic and wild animals*. Academy of Sciences of the USSR, Vladivostok, USSR, 84–88 (in Russian).
- Demshin N.I. 1968. Sexually mature trematode *Hirudineatrema oschmarini* sp. gen. et subfam. nov. – a parasite of leeches. *Proceedings of the Far Eastern Branch of the Academy of Sciences of the USSR*, 26, 41–45 (in Russian).
- Demshin N.I., Leonov V.A. 1975. Oligochaetes and leeches as intermediate hosts of helminths. Nauka, Novosibirsk, USSR, 189 pp. (in Russian).
- Demshin N.I. 1976. Larval development of the cestode *Sacciuterina stellifera* (Dilepididae). *Zoologicheskij Zhurnal*, 55, 17–22.
- Demshin N.I. 1985. Postembryonic development of the cestode *Nippotaenia mogurndae* (Nippotaeniidea, Nippotaeniidae). *Parazitologiya*, 19, 39–43.
- Fish T.D., Vande Vusse F.J. 1976. *Hirudicolotrema richardsoni* gen. et. sp. n. (Trematoda: Macroderoididae) from Minnesota hirudinid leeches. *Journal of Parasitology*, 62, 899–900. DOI: <http://dx.doi.org/10.2307/3279181>.
- Font W.F. 1980. The effect of progenesis on the evolution of *Alloglossidium* (Trematoda, Plagiorchiida, Macroderoididae). *Acta Parasitologica Polonica*, 27, 173–183.
- Font W.F. 1994. *Alloglossidium greeri* sp. nov. (Digenea: Macroderoididae) from the cajun dwarf crayfish, *Cambarellus schufeldti*, in Louisiana, U.S.A. *Transactions of the American Microscopical Society*, 113, 86–89. DOI: <http://dx.doi.org/10.2307/3226583>.
- Font W.F., Lotz J.M. 2008. Family Macroderoididae McMullen, 1937. In: (Eds. D.I. Gibson, R.A. Bray, A. Jones). *Keys to the Trematoda*. Vol. III. CABI Publishing and the Natural History Museum, London, U.K., pp. 373–380.
- Klemm D.J. 1982. Leeches (Annelida: Hirudinea) of North America. Environmental Monitoring and Support Laboratory, Office of Research and development, U.S. Environmental Protection Agency, Cincinnati, Ohio, USA, 177 pp.
- Neumann M.P., Vande Vusse F.J. 1976. Two new species of *Alloglossidium* Simer 1929 (Trematoda: Macroderoididae) from Minnesota leeches. *Journal of Parasitology*, 62, 556–559. DOI: <http://dx.doi.org/10.2307/3279414>.
- Oshmarin P.O., Demshin N.I., Lebedev B.I. 1970. Helminths of animals of South-East Asia. Moscow, Nauka, USSR, 220 pp.
- Schmidt G.D., Chaloupka K. 1969. *Alloglossidium hirudicola* sp. n., a neotenic trematode (Plagiorchiidae) from leeches, *Hae-mopis* sp. *Journal of Parasitology*, 55, 1185–1186. DOI: <http://dx.doi.org/10.2307/3277253>.
- Smythe A.B., Font W.F. 2001. Phylogenetic analysis of *Alloglossidium* (Digenea: Macroderoididae) and related genera: life-cycle evolution and taxonomic revision. *Journal of Parasitology*, 87, 386–391. DOI: <http://dx.doi.org/10.2307/3285056>.
- Timmers S.F. 1979. *Alloglossidium schmidtii* sp. n. (Trematoda: Macroderoididae) from hirudinid leeches in Manitoba. *Proceedings of the Helminthological Society of Washington*, 46, 180–184.
- Tkach V.V., Mills A.M. 2011. *Alloglossidium fonti* sp. nov. (Digenea, Macroderoididae) from black bullheads in Minnesota with molecular differentiation from congeners and resurrection of *Alloglossidium kenti*. *Acta Parasitologica*, 56, 154–162. DOI: <http://dx.doi.org/10.2478/s11686-011-0025-y>.
- Vande Vusse F.J. 1980. Revision of *Alloglossidium* Simer 1929 (Trematoda: Macroderoididae) and description of *A. microspinatum* sp. n. from a leech. *Journal of Parasitology*, 66, 667–670. DOI: <http://dx.doi.org/10.2307/3280527>.