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Lucionema balatonense gen. et sp. n., a new nematode of a new family Lucionematidae fam. n. (Dracunculoidea) from the swimbladder of the European pikeperch, *Stizostedion lucioperca* (Pisces)

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Abstract. A new nematode genus and species, *Lucionema balatonense* gen. et sp. n., is described from the swimbladder of the European pikeperch, *Stizostedion lucioperca* (L.), from Lake Balaton in Hungary; a new dracunculoid family Lucionematidae fam. n. is established to accommodate it. The hitherto monotypic family Lucionematidae shows affinities with the families Skrjabillanidae and Daniconematidae, differing from them mainly in having simple oesophagus without external oesophageal glands and the vulva situated near the middle of body; from the first family also in the absence of the buccal capsule and the bursa-like caudal alae in the male. The genus *Lucionema* gen. n. is characterized mainly by the presence of 8 cephalic papillae in two circlets, absence of spicules, presence of the copulatory plate, only 2 pairs of postanal papillae in the male, and by the distal part of the monodelphic uterus forming a posteriorly directed coil. The body length of *L. balatonense* females is 1074-1782 μm, that of the only available male 770 μm. A key to the families of the Dracunculoidea is presented.

During recent studies on the parasites of fish in Lake Balaton in Hungary, carried out by the junior authors of this paper (K. Molnár and Cs. Székely), a previously undescribed dracunculoid nematode was found in the swimbladder of the European pikeperch, *Stizostedion lucioperca* (L.). Since this parasite species exhibits unique morphological features, being considerably different from members of the related families Skrjabillanidae and Daniconematidae, creation of an independent genus *Lucionema* gen. n. and a new family Lucionematidae fam. n. is proposed.

MATERIALS AND METHODS

During a three-year survey on Lake Balaton fishes in Hungary from April 1994 to March 1997, 160 specimens of the European pikeperch, *Stizostedion lucioperca* (L.), were examined for parasitic infections. The total length of fish seined by commercial fishermen varied between 16 to 43 cm. They were transported alive to the laboratory of the Veterinary Medical Research Institute in Budapest where they were subsequently examined within a few days. After killing the fish with a blow on the head, a complete parasitological dissection including all organs was made. The minced tissue of the swimbladder was placed in a petri dish with a small amount of physiological saline (0.65%) and was thoroughly examined under the dissecting microscope, using fine mounting needles. Both live and fixed specimens of the adult and larval stages were video-recorded, and photos were taken from digitalized pictures with the help of a video-image program. The nematodes were first washed in physiological saline and then fixed in hot (80°C) mixture of saline and 40% formalin (9 : 1). The only recovered male and one gravid female were video-screened. Unfortunately, during later manipulation, the male was lost and, therefore, only its record on the video-tape was used for the description. The females were cleared with glycerine for examination. Drawings were made with the aid of an Aristoplan microscope drawing attachment. A few females were used for scanning electron microscopy (SEM). For SEM, nematodes from 4% formaldehyde were postfixed in 1% OsO_4, dehydrated through an ethanol series to acetone and subjected to critical point drying. Specimens were coated with gold and examined with a JSM-6300 scanning electron microscope at an accelerating voltage of 15 kV. However, the results were not satisfactory and, consequently, the SEM micrographs were used only for the reconstruction of the cephalic end in an apical view (Fig. 1C). All measurements are given in μm unless otherwise stated. The specimens have been deposited in the helminthological collection of the Institute of Parasitology, Academy of Sciences of the Czech Republic, in České Budějovice (Cat. No. N-689) and in the Veterinary Medical Research Institute, Hungarian Academy of Sciences, in Budapest.

RESULTS

In a 37 cm long pikeperch, *Stizostedion lucioperca* (L.), collected in September 1995, nematode larvae of an unknown species resembling skrjabillanid nematodes were found under the serosa of the swimbladder. After the first finding, 72 further pikeperch specimens were examined, but only 4 of them proved to be infected by larval nematodes. The intensity of infection in these fish was 20, 32, 23 and 1, respectively. Only a single

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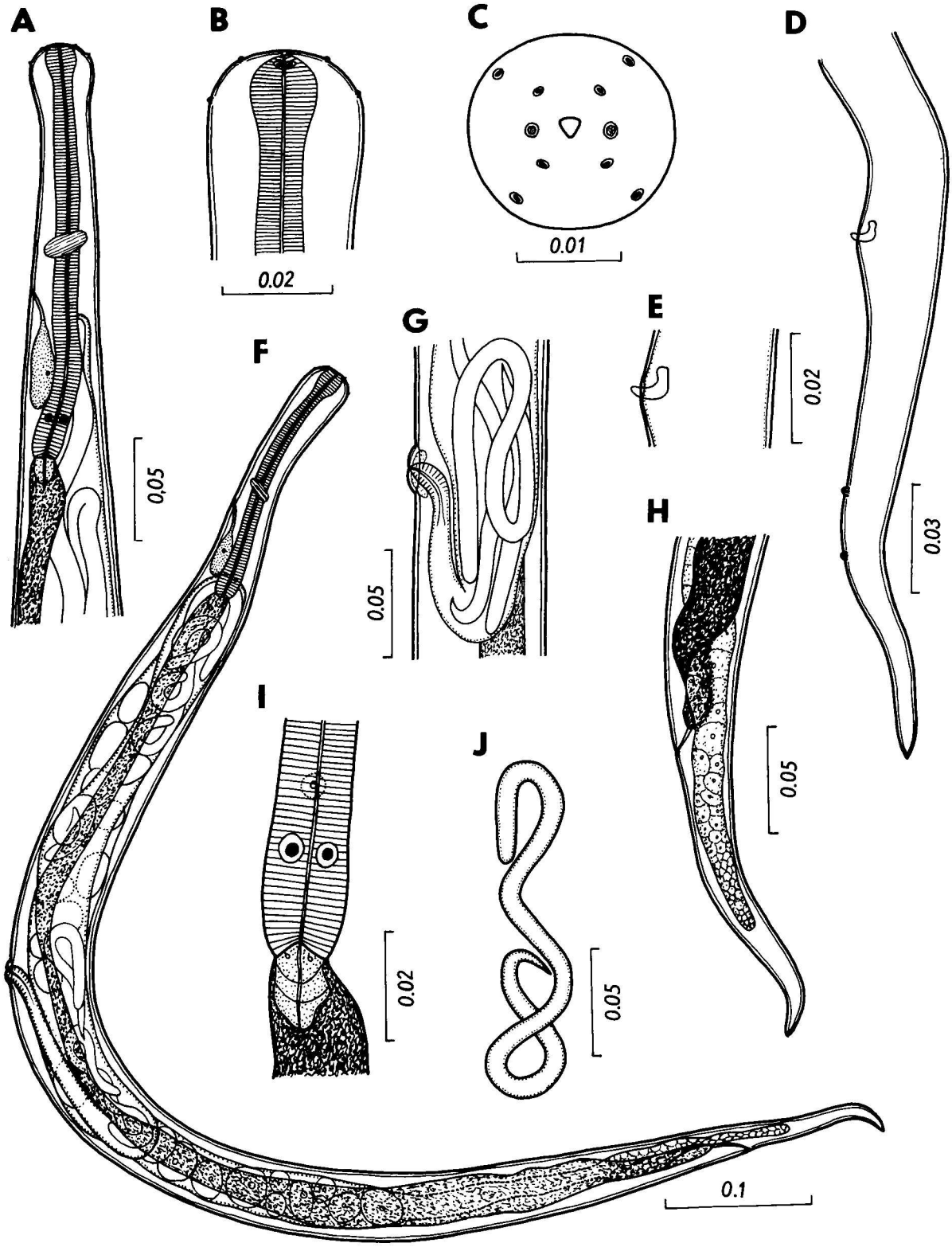

Fig. 1. *Lucionema balatonense* sp. n. A – anterior part of body of gravid female; B, C – cephalic end of female, lateral and apical views (C – reconstruction based on SEM micrographs); D – caudal end of male; E – region of cloacal opening with copulatory plate; F – general view of gravid female; G – region of vulva; H – caudal end of female; I – junction of oesophagus and intestine in female; J – larva from uterus. Scale bars in millimetres.

pikeperch, 38 cm long, harboured undoubtedly conspecific adult nematodes.

An infection of so far unidentified skrjabillanid-like larvae, supposed to belong to this species (at least part

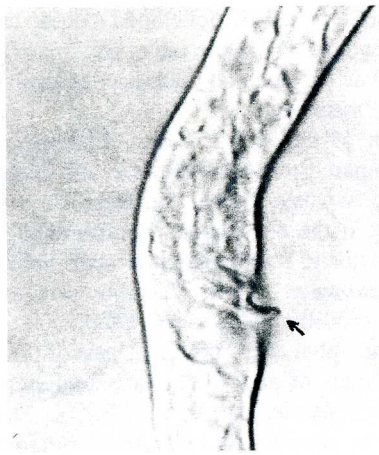

Fig. 2. *Lucionema balatonense* sp. n. Region of cloaca in male with distinct copulatory plate (arrow) (× 1000); photo taken from digitalized picture with help of video-image program.

of them) was found in the haemocoel of fish louse, *Argulus foliaceus* (L.), in the specimens collected from the fins of some randomly selected pikeperch specimens. Seventeen out of the 37 fish louse specimens examined proved to be infected by larvae of different size, moving vigorously in the crustacean intermediate host. Skrjabillanid-like larvae from *Argulus* and fish in Lake Balaton will be the subject of the following study.

Lucionema balatonense gen. et sp. n. Figs. 1,2

Description: Very small, fusiform whitish nematodes with thin, almost smooth cuticle. Cephalic end rounded. Oral aperture small, triangular, surrounded by eight small submedian cephalic papillae arranged in two circlets and pair of rather large circular lateral amphids. Oesophagus simple, muscular throughout, without any external oesophageal glands; its anterior end somewhat inflated, posterior part moderately distended, provided with three large cell nuclei. Oesophagus opening into intestine through large valve. Nerve ring encircling oesophagus approximately at its mid-length, excretory pore somewhat posterior to level of nerve ring. Deirids not found. Tail of both sexes conical, long, with pointed tip.

Male (based on 1 specimen recorded on video-tape): Length of body 770, maximum width 21. Oesophagus 83 long, width at level of its anterior inflation 14. Nerve ring 63 from anterior extremity. Tail conical, elongate, 94 long, ending in blunt point. Only 2 pairs of subventral postanal papillae present, being situated approximately at mid-length of tail; preanal papillae absent. Unpaired, bent, well sclerotized copulatory plate present; its length (distance between its proximal and distal ends) 8 in lateral view (Fig. 2).

Female (based on 10 specimens; measurements of holotype in parentheses): Length of body of gravid females with larvae in uterus 1074-1782 (1360), maximum width 39-72 (69) at middle of body. Anterior, oesophageal part of body narrowed. Cephalic end slightly inflated, 27-45 (39) wide; width at narrowest part of neck region at level of nerve ring 24-36 (33). Oesophagus 204-255 (210) long, representing 14-19% (15%) of body length, its maximum width at posterior part 15-21 (18). Nerve ring and excretory pore 105-120 (105) and 132-192 (135), respectively, from anterior extremity. Light-coloured intestine narrow at its anterior part and relatively broad at posterior part. Rectum short hyaline tube. Anus functional. Tail simple, conical, 96-159 (114) long, with pointed tip. Reproductive system monodelphic. Single ovary at region of posterior end of intestine, extending posteriorly to about middle of tail or more. Oviduct narrow, rather long; uterus running anteriorly to about level of end of oesophagus, then it returns back to certain distance posterior to vulva; distal part of uterus again oriented anteriorly and gradually turning into short muscular, posteriorly directed vagina. In the direction from its distal to proximal end, uterus containing gradually fully developed larvae, developing embryos and eggs. Vulva near middle of body, being slightly preequatorial (at 40–46% (43%) of body length), 476-748 (558) from anterior extremity. Vulvar lips somewhat elevated. Larvae from uterus with rounded cephalic end and pointed tail; their body length 240-330 (240), maximum width 12 (12).

Type host: European pikeperch, *Stizostedion lucioperca* (L.) (Percidae, Perciformes).

Site of infection: Swimbladder wall.

Type locality: Lake Balaton, Hungary (holotype and all paratypes collected on 3 October 1996).

Prevalence: 1.4% for adult nematodes; 5.4% for larvae.

Intensity: 29 for adult nematodes; 1-32 (mean 16) for larvae.

Deposition of types: Institute of Parasitology, Academy of Sciences of the Czech Republic, České Budějovice (Helm. Coll. No. N-689, holotype and 10 paratypes) and Veterinary Medical Research Institute, Hungarian Academy of Sciences, Budapest (10 paratypes).

Etymology: The generic name *Lucionema* is composed of two parts: *Lucio* and *nema*; the former is the anterior part of the scientific name of the host fish (*lucioperca*), whereas the latter means the nematode. The specific name *balatonense* relates to the place of its origin, i.e. Lake Balaton.

Genus ***Lucionema*** gen. n.

Diagnosis: Lucionematidae. Body fusiform, head end rounded, provided with eight cephalic papillae arranged in two circlets and pair of lateral amphids. Buccal capsule absent. Oesophagus undivided, with bulb-like inflation at its anterior end. Nerve ring near mid-length of oesophagus, excretory pore situated slightly posterior to nerve ring level. Tail of male conical, caudal alae absent; two pairs of postanal papillae present. Copulatory plate present, spicules absent. Female with functional anus; female tail conical, pointed. Vulva near middle of

body. Uterus filled with larvae extending anteriorly to posterior end of oesophagus. Parasites of swimbladder of fishes.

Type– and the only species: *Lucionema balatonense* sp. n.

Family Lucionematidae fam. n.

Diagnosis: Dracunculoidea. Body fusiform. Head end with eight papillae. Buccal capsule absent. Oesophagus undivided. Tail of male long, without caudal alae, provided with few pairs of postanal papillae; preanal papillae absent. Sclerotized copulatory plate present, spicules absent. Vulva near middle of body. Monodelphic. Viviparous. Parasites of fishes.

Type– and the only genus: *Lucionema* gen. n.

DISCUSSION

The general morphology of *Lucionema balatonense* shows that this nematode species belongs to the superfamily Dracunculoidea Stiles, 1907 in the conception of Chabaud (1975); it is indicated mainly by the morphology of the cephalic end, structure of the oesophagus, character of genital organs in the female, presence of a copulatory plate in the male, viviparity, and also by the fact that these nematodes are tissue-dwelling parasites.

According to Moravec and Køie (1987), the superfamily Dracunculoidea includes seven families: Dracunculidae Stiles, 1907, Micropleuridae Baylis et Daubney, 1926, Philometridae Baylis et Daubney, 1926, Guyanemidae Petter, 1975, Anguillicolidae Yamaguti, 1935, Skrjabillanidae Shigin et Shigina, 1958 and Daniconematidae Moravec et Køie, 1987. Of them, *L. balatonense* appears to be most closely related with the families Skrjabillanidae and Daniconematidae, but its affinities with Anguillicolidae are also apparent.

A characteristic feature of *L. balatonense* is the presence of the sclerotized copulatory plate in the male, which apparently substitutes the function of missing spicules. Within Dracunculoidea, this organ occurs only in the Skrjabillanidae (well developed in *Skrjabillanus* Shigin et Shigina, 1958 and *Molnaria* Moravec, 1968, weakly developed in *Esocinema* Moravec, 1977 and absent in *Sinoichthyonema* Wu in Chen, 1973) and the Daniconematidae (well developed in *Mexiconema* Moravec, Vidal et Salgado Maldonado, 1992 and absent in *Daniconema* Moravec et Køie, 1987) (Moravec et al. 1992, Moravec 1994); both these families include fish parasites.

However, in contrast to *L. balatonense*, the oesophagus in species of Daniconematidae is divided into an anterior muscular and a posterior glandular parts, whereas in species of Skrjabillanidae it is muscular throughout, but provided with a large external, posteriorly extending oesophageal gland. The structure of the oesophagus is generally considered a very important feature in nematode taxonomy. Moreover, all skrjabillanids possess a well developed buccal capsule and bursa-like caudal alae in the male, which are absent in *Lucionema*. The capsule and male caudal alae are absent in Daniconematidae.

The situation of the vulva is also different in *Lucionema* as compared to Skrjabillanidae and Daniconematidae: in the two last named families, the vulva is always anterior, at the oesophageal region of the body or slightly posterior to it, whereas it is near the middle of body in *Lucionema*. In this, *Lucionema* somewhat resembles the Anguillicolidae (*Anguillicola* Yamaguti, 1935) having the vulva at the posterior part of the body; also the oesophagus of anguillicolids is undivided and they are the swimbladder parasites of fish as *Lucionema*. However, in contrast to *Lucionema*, *Anguillicola* species have a large buccal capsule armed with peribuccal teeth, their uterus is amphidelphic and the male has preanal papillae in addition to other differentiating features.

Since *L. balatonense* markedly differs in important taxonomic features from members of both closely related families Skrjabillanidae and Daniconematidae, we consider it necessary to create for it a new independent family Lucionematidae fam. n. Differences between this family and other families of Dracunculoidea are more obvious from the key at the end of this paper.

The discovery of this nematode parasite is quite unique, because the helminth fauna of freshwater fishes in Europe has been studied by helminthologists for more than two centuries and Europe is the best explored continent in this respect (Moravec 1994). However, the fact that the parasite remained undiscovered for such a long time could be explained by its very small measurements, its susceptibility to osmotic pressure and its hidden way of life in the wall of the swimbladder. It can be supposed that *L. balatonense* is indigenous to Europe and that subsequent studies may show its occurrence in other European localities. Such small nematodes pass unnoticed during routine examinations of fish and, on the contrary, a thorough examination of all body organs may result in surprising findings like that of *L. balatonense* or the recently discovered *Skrjabillanus cyprini* Molnár et Moravec, 1997 from the base of scales of the European common carp, *Cyprinus carpio* L. (Molnár and Moravec 1997).

In our survey the pikeperch showed a very low level of infection with *L. balatonense*. This can be contributed to a low prevalence of this parasite in the pikeperch but it cannot be excluded that the swimbladder is only one of the possible locations for this histozoic parasite. In low-intensity infection larval and adult specimens may be located in the tissues of other organs which are more difficult to study than the swimbladder. Data obtained by Molnár and Moravec (1997; unpublished data) indicate that in the scales, in the abdominal cavity and under the skin some more, hitherto unknown species of skrjabillanid nematodes can be found in

different fishes. A more common occurrence of this species is supported by the observation that about half of the *Argulus foliaceus* specimens collected at random from pikeperch proved to be infected by skrjabillanid-like nematodes. It is highly probable that *L. balatonense* follows the same pattern of development as skrjabillanids (see Tikhomirova 1970), which means that the first-stage larvae released from the viviparous nematodes are taken up by the intermediate host, *Argulus* spp., while sucking on infected pikeperch and third-stage larvae are inoculated into other pikeperch specimens. The relatively high infection of *Argulus* specimens collected from the fins of randomly selected pikeperch, however, is only an inadequate proof of the more frequent occurrence of this nematode in fish, because this intermediate host can change fish hosts during its life and larvae of different skrjabillanid-like species can develop inside its body.

Key to families of the Dracunculoidea

1 Buccal capsule present; spicules absent 2
– Buccal capsule absent or reduced to a peribuccal ring; spicules present or absent 3
2 Body fusiform; tail of male without caudal alae; sclerotized copulatory plate in male absent; vulva posterior; parasites of swimbladder of eels ..... **Anguillicolidae**
– Body filiform; tail of male with large caudal alae in form of a bursa; sclerotized copulatory plate in male present or absent; parasites of peritoneal cavity of palaearctic freshwater fishes .... **Skrjabillanidae**
3 Vulva anterior or preequatorial, well developed in mature female; monodelphic; peribuccal ring absent; anus functional 4
– Vulva equatorial or posterior, more or less completely atrophied in mature female; two ovaries present (except *Phlyctainophora*) 6
4 Oesophagus undivided, glandular part of oesophagus absent; vulva preequatorial, near middle of body; uterus extending anteriorly far anterior to level of vulva; spicules and caudal alae absent, copulatory plate present ..... **Lucionematidae** fam. n.
– Oesophagus divided into muscular and glandular parts; vulva distinctly anterior, usually at oesophagus level; uterus not exceeding anteriorly level of vulva; spicules, copulatory plate and caudal alae present or absent 5
5 Spicules present; caudal alae in male present ..... **Guyanemidae**
– Spicules absent; caudal alae in male absent ..... **Daniconematidae**
6 Glandular part of oesophagus short or absent; tail of male rounded and very short (except *Philonema*); peribuccal ring absent; parasites of fishes ..... **Philometridae**
– Glandular part of oesophagus long; tail of male sharply pointed, of average length 7
7 Peribuccal ring absent; sexual dimorphism moderate; anus functional; parasites of archaic reptiles (crocodilians and chelonians) ..... **Micropleuridae**
– Peribuccal ring present (except *Avioserpens*); sexual dimorphism marked; anus atrophied in adult; parasites of reptiles, birds, and mammals ..... **Dracunculidae**

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