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HISTOLOGICAL CHANGES IN THE SWIMBLADDER WALL OF EELS DUE TO ABNORMAL LOCATION OF ADULTS AND SECOND STAGE LARVAE OF *Anguillicola crassus*

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In severe *Anguillicola crassus* infection of eels, adult helminths and 2nd stage larvae staying in the swimbladder lumen may occasionally get, through minor lesions of the tunica interna, into the subserosa of the swimbladder wall where they die and disintegrate. A thin connective tissue capsule is formed around the helminths that behave as foreign bodies in intercellular location, while the lacunas of the surrounding loose connective tissue comprise melanin-containing macrophages. In the environment of the 2nd stage larvae the formation of giant cells is a typical finding.

Key words: *Anguillicola crassus,* Nematoda, eel, swimbladder, abnormal location, histopathology

Since *Anguillicola crassus* was introduced into Europe in the mid-1980s, (Neumann, 1985) numerous papers have dealt with its prevalence (Peters and Hartmann, 1986; Taraschewski et al., 1987; Hartmann, 1987; Dupont and Petter, 1988; Belpaire et al., 1989; Koops and Hartmann, 1989; Kennedy and Fitch, 1990; Koie, 1991; Székely et al., 1991; Moravec, 1992), life cycle (De Charleroy et al., 1990; Haenen and van Banning, 1991; Höglund and Thomas, 1992; Thomas and Ollevier, 1992), seasonal occurrence (van Willigen and Dekker, 1989), and pathogenic effect exerted on the host (Mellergaard, 1988; Haenen et al., 1989; Boon et al., 1989; Boon et al., 1990*a,b,c*; van Banning and Haenen, 1990; Molnár et al., 1991; Möller et al., 1991; Sprengel and Lüchtenberg, 1991; Höglund et al., 1992). At the same time, data on the histopathological changes caused by the parasite can be found only in the works of Haenen et al. (1989), van Banning and Haenen (1990), and Molnár et al. (1993). The latter authors gave a detailed description of the general lesions produced by adult worms and by larvae in the mucous membrane of the swimbladder and in the subserosa, but only incidentally mentioned the mechanical injury caused to the swimbladder wall. At the same time, Liewes and Schaminee-Main (1987) and Kamstra (1990) graded by severity the swimbladder lesions caused by anguillicolosis in eels. They regarded

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as the most severe lesion the rupture of the swimbladder wall and the appearance of a brownish-blackish substance consisting of nematode debris in the swimbladder wall that had been replaced by a thick layer of connective tissue.

This paper presents the histopathological changes resulting from the anguillicolosis-induced discontinuity of the swimbladder wall, and provides data on the histogenesis of the granuloma-like lesions caused by helminths and larvae abnormally entering the swimbladder wall, as well as on the nature of the pigmentation which is seen in some cases.

Materials and methods

The material used in this study comprised eels that had been derived from Lake Balaton and dissected in 1992 (Molnár et al., 1993). In 1993, complementary studies were carried out: of 344 eels caught from different regions of Lake Balaton, only those specimens which exhibited changes resulting from swimbladder lesions (helminths and 2nd stage larvae located in the swimbladder wall, or intensive pigment formation) were processed for histology. In contrast to the earlier paper, here we do not follow Dorn's nomenclature, and refer to the loose connective tissue surrounded by the serosa and the muscular layer by the name of subserosa, as part of the tunica externa, rather than by the name of submucosa. According to the classification adopted by us, the tunica interna comprises the muscular layer and the mucosa.

Swimbladders intended for histological processing were placed into Bouin's solution in their entirety for some minutes, then were cut through at the affected part or transversely in the middle, and the smaller parts were again fixed in Bouin's solution for 4 hours. The materials were embedded in paraffin wax and cut into 4 μm thick sections. The preparations were stained with haematoxylin-eosin for general information, with picrosirius stain to study collagenic fibres, by Brown-Brenn and by Ziehl-Neelsen for bacteria, by Perls for haemosiderin, and by the periodic acid–Schiff (PAS) reaction for mucous cells. Some sections were treated with 10% H_2O_2 solution for one hour, then stained also by the method of Perls and by the Oil-red procedure.

Electron microscopic examination involved the subsequent processing of material that had been previously fixed in Bouin's solution and embedded in paraffin wax.

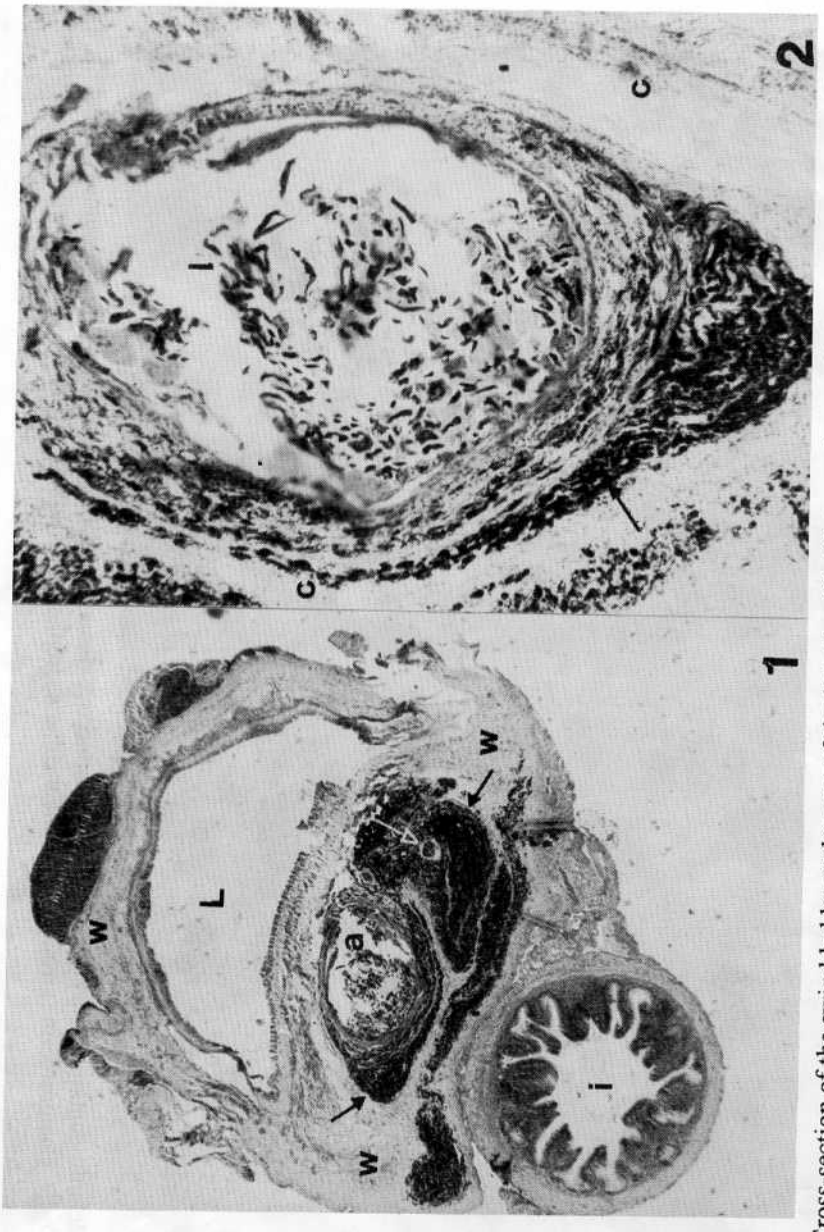

Fig. 1. Cross-section of the swimbladder and a part of the intestine (i). Note an abnormally located female *Anguillicola crassus* (a) in the wall (w) of the swimbladder. The worm was translocated here from the lumen (L) due to the rupture of the muscular layer and mucosa. An adult (a) and a 4th stage larva (empty arrow) are surrounded by melanomacrophages (arrow). Haematoxylin and eosin (H. and E.), × 35

Fig. 2. Enlarged picture of the extraluminal *A. crassus* female shown in Fig. 1. Second stage larvae (l) fill the body of the helminth. The connective tissue of subserosa (c) around the worm is infiltrated by melanomacrophages (arrow). H. and E., × 130