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Authors: Poole, B. C., and Dick, T. A.

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## LIVER PATHOLOGY OF YELLOW PERCH, *PERCA FLAVESCENS* (MITCHILL), INFECTED WITH LARVAE OF THE NEMATODE *RAPHIDASCARIS ACUS* (BLOCH, 1779)

B. C. Poole and T. A. Dick

Department of Zoology, University of Manitoba, Winnipeg, Manitoba R3T 2N2, Canada

**ABSTRACT:** Larvae of the nematode *Raphidascaris acus* were found free or encapsulated in the liver of yellow perch. Blood vessels were distorted or destroyed during larval migrations and larvae were eventually encapsulated in a thick-walled whitish nodule. Successful walling-off of the parasite resulted in the formation of a collagenous nodule and a complete loss of the worm. No mortality of perch was associated with larval *R. acus* but the introduction of susceptible fishes into a lake harboring this parasite may be important in some stocking programs.

### INTRODUCTION

*Raphidascaris acus* is a widely distributed parasite occurring in the liver of fishes from Eurasia and North America (Moravec, 1970; Hoffman, 1967; Margolis and Arthur, 1979). This nematode is known to cause severe emaciation, complete loss of function of infected organs, and death of heavily infected fish such as bream, *Abramis brama* (L.) (Osmanov, 1953, in Petrushevski and Shulman, 1961), but such damage may not be typical. Although the pathogenesis is not well understood, Logachev and Pronina (1975) described liver capsules enclosing larvae of *R. acus* from sand sculpin, *Paracottus kessleri* Dybowskii, collected in the USSR. Yellow perch is one of the main natural intermediate hosts of *R. acus* in North America, at least for the larval stages in the liver, but no information is available on pathology induced by larval movement or encapsulation. This is particularly important to know as there is no evidence to date that *R. acus* causes severe damage or mortality in perch. The objective of this study was to describe the lesions associated with *R. acus* infections in the liver of naturally infected yellow perch.

### MATERIALS AND METHODS

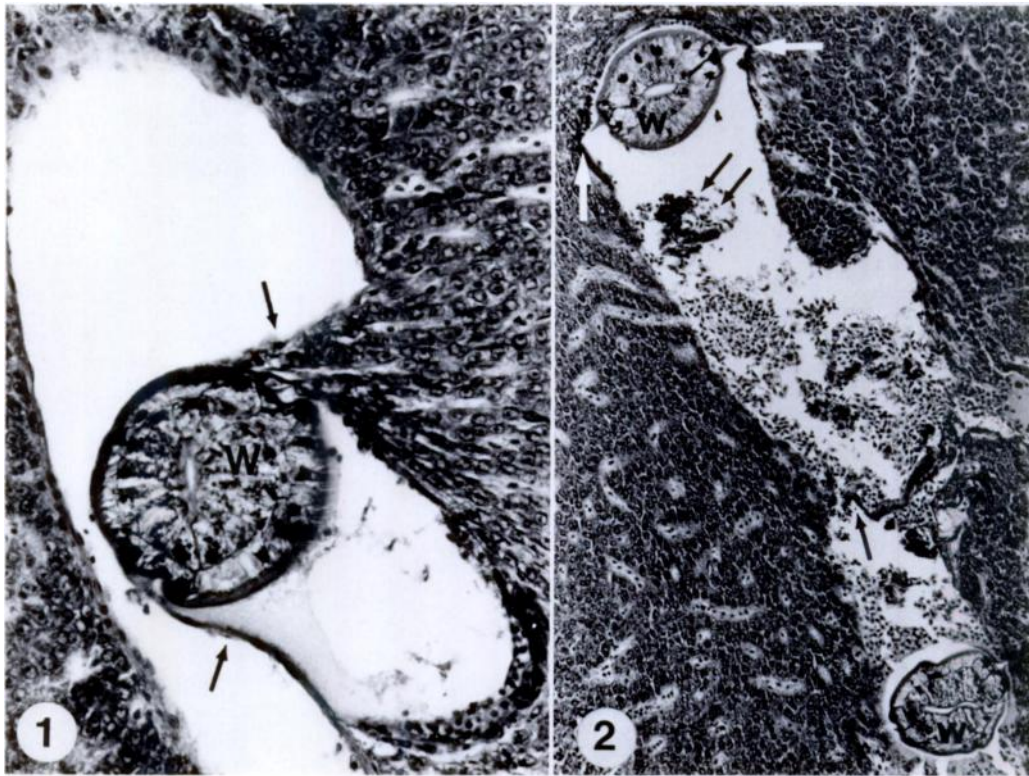
Perch were collected with gill and trap nets from June to October of 1982 from Quigly Lake, Manitoba in north-central Canada (54°53'N, 101°07'W). Swedish experimental nets 2 m deep and consisting of six 10-m panels (10, 19, 35, 45, 55, and 65 mm, measurements on the bar) were all bottom sets in 2-6 m of water. Forty whole perch were kept fresh on ice and examined within 36 hr of capture. Pieces of infected liver (ranging from 25 to 50% of whole liver) or whole livers with typical white pustules on the surface were removed from 20 perch, fixed in Bouin's, embedded in paraffin, and serially sectioned at 10 µm. Sections were stained with either Gill's hematoxylin and eosin for cellular detail, or with picro-sirius red to indicate the presence of collagen. Whole livers from 440 perch in this area were also examined with the aid of a trichinoscope under a Wild M3 dissecting microscope for larvae of *R. acus*.

### RESULTS

Larvae of *R. acus* were found free or encapsulated in the livers of perch. Unencapsulated larvae were found in the major hepatic blood vessels and aside from slight hemorrhaging there were no lesions. No free larvae were observed in liver parenchyma. Tissue damage was more apparent as larvae grew and migrated within the blood vessels. Hepatic cords were displaced as worms coiled and migrated and the walls of the vessels were distorted (Fig. 1). Mechanical pressure by lateral alae may have accounted for some of the dam-

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FIGURES 1, 2. Non-encapsulated larvae of *Raphidascaris acus* in the liver of yellow perch. 1. Cross section of a migrating worm (w) stained with hematoxylin and eosin (H&E). Note distorted hepatic cords and blood vessel walls (arrows).  $\times 328$ . 2. The destruction of blood vessels by migrating worms (w) and the formation of one large blood sinus. Note the distortion of blood vessel walls by the lateral alae region of the worm (white arrows), liver parenchymal debris in the lumen of the blood vessel (double black arrows), and the destruction of blood vessel walls (single black arrow). H&E,  $\times 148$ .

age and the eventual breakdown of vessel walls (Fig. 2). In contrast, areas of blood vessels not associated with the lateral alae region of larvae remained undamaged.

Encapsulated larvae of *R. acus* appeared normal and were distributed in close proximity to blood vessels throughout the liver, but were found frequently near the periphery where the capsules were visible to the eye as small white pustules. Only one larva was found in each of the capsules examined by serial sectioning or with the aid of a trichinoscope. The wall of the capsule was cellular in appearance when stained with hematoxylin and eosin, with fibroblasts being the main cellular component (Figs. 3, 6). Strongly

eosin-positive granulocytes were frequently observed scattered among the fibroblasts. Large vacuolated cells, some with eccentric nuclei, were also associated with the host capsule, but in close proximity to the larva (Fig. 6). These cells were identified as macrophages and/or degenerating hepatocytes. Accumulations of leucocytes were found often along the outer wall of the capsule (Fig. 3). A picrosirius red stain revealed extensive collagen in the capsule wall, deposited either as tight whorls (Fig. 4) or as loosely aggregated rings (Fig. 5).

The center of the capsule contained degenerating red blood cells, leucocytes, hepatocytes (Fig. 3) and amorphous mate-

rial (Fig. 5). Necrotic tissue resembling this amorphous material was commonly observed in the intestinal lumen of encapsulated larvae, but was never seen in the intestine of free larvae. Figure 5 suggests that the worm attempted to move away from the original encapsulation but was eventually re-encapsulated. No capsules with degenerate larvae were observed during either the trichinoscope examination or from serial sectioning. The final stage of capsule formation (Fig. 7) was the absence of worms and a well-defined collagenous nodule with a vacuolated and necrotic center.

No mortality of perch was associated with infections by larval *R. acus*.

#### DISCUSSION

The presence of free larvae of *R. acus* in the blood vessels of the liver of perch appears to be related to worm size and perhaps to the stage of the parasite. This is supported by other studies where the length of time which larvae of *R. acus* remained unencapsulated in the liver ranged from 153 days post-infection in the European stone loach, *Neomacheilus barbatus* (L.) (Moravec, 1970) to almost a year (autumn until the following summer) in yellow perch (Smith and Anderson, 1982). Larvae are encapsulated as a result of a strong cellular response. It is not clear what initiates capsule formation, but damage to blood vessels and adjacent tissues by these larvae could elicit a strong cellular response and subsequent encapsulation. Although the leucocytes associated with encapsulated larvae of *R. acus* in this study were not identified as polymorphonuclear leucocytes, the latter cells are involved in the production of suppurative exudate reactions (Roberts, 1978). Logachev and Pronina (1975) reported fibroblasts and extensive collagen surrounding larvae of *R. acus* infecting the liver of sand sculpin in Russia, and occasionally found blood vessels penetrating the capsule wall. Although blood vessels were not observed

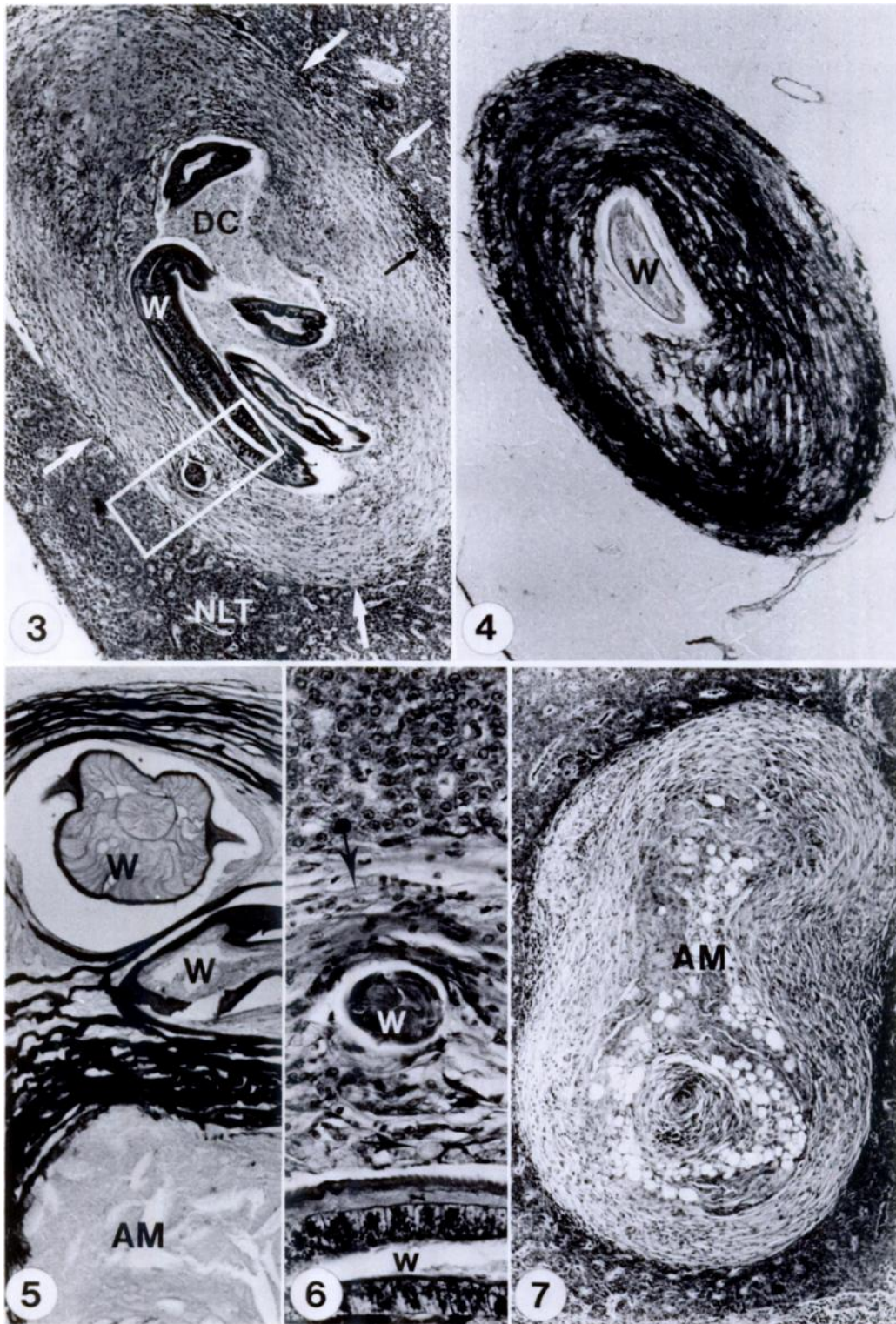
in the capsule wall in this study, degenerating blood cells were found in the lumen of the capsule.

The presence of necrotic cells in the intestinal lumen of encapsulated larvae resembling the necrotic tissue in the center of the host capsule was also observed by Logachev and Pronina (1975), and suggests that these worms may feed on this material. The absence of blood or any other tissue from the intestinal lumen of migrating larvae of *R. acus* suggests that they may not feed until they are encapsulated by the intermediate fish host.

It is generally believed that only second- and third-stage larvae of *R. acus* are present in fish livers (Moravec, 1970; Smith and Anderson, 1982) and we assume most lesions and encapsulations were induced by these stages. However, the presence of two encapsulated sexually mature male worms recovered from the liver of perch (unpubl. data) suggests that other stages may be involved in the formation of lesions, even if rarely. Multiple encapsulations of worms were noted by Moravec (1970) and Logachev and Pronina (1975) but only one worm was found in each of the capsules examined in our study.

The numbers of *R. acus* in livers of perch in this study were observed to increase with the length of the host, but there was no effect on overall fish growth (unpubl. data). However, Osmanov (1953, in Petrushevski and Shulman, 1961) reported that heavy infections of *R. acus* (up to 1,035 larvae) in the liver and intestinal walls of bream caused organ malfunction and the death of the fish host. These differences may be related to the numbers of larvae of *R. acus* in the livers of fishes and the species of fish infected. The small portion of the liver (5%) occupied by the parasite and capsule and the absence of multiple worm encapsulations in perch may decrease the potential for severe damage. However, in light of the reports on mortality of some fishes in the USSR we should be somewhat cautious about the





introduction of more susceptible fishes to aquatic systems with well-established populations of *R. acus*.

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FIGURES 3–7. Encapsulated larvae of *Raphidascaris acus* in the liver of yellow perch. 3. Typical capsule surrounding a worm (w) within normal liver tissue (NLT). Note the mass of degenerating cells (DC) in the center of the capsule and the accumulation of leucocytes (black arrow). White arrows indicate the boundaries of the capsule. (See Fig. 6 for magnification of boxed area.) H&E,  $\times 77$ . 4. The successful walling-off of a worm (w) with extensive collagen deposition (black whorls). Note the loss of degenerating cells in the capsule center. Picro-sirius red (PSR),  $\times 80$ . 5. The network of collagen (black whorls) in a capsule where the worm (w) appears to have moved away from the initial host response where only amorphous material (AM) remains. PSR,  $\times 330$ . 6. Enlargement of a portion of capsule wall (see boxed area in Fig. 3) showing macrophages (arrow) and fibroblasts (arrow with dot). H&E,  $\times 318$ . 7. Late stage capsule composed of collagen and interspersed amorphous material (AM) and no worm. H&E,  $\times 99$ .