Pathological effect on naturally infected kidneys of freshwater murels by *Euclinostomum heterostomum* metacercariae

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Received: 15 December 2013 / Accepted: 16 April 2014 / Published online: 6 May 2014
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Abstract Present investigation was carried out to study the histo-pathological alterations induced by metacercariae of digenetic trematode, *Euclinostomum heretostomum* on the kidney of *Channa striatus* and *Channa marulius* in natural condition. Histo-architectural alterations induced by parasite included reduced size of glomeruli, severe degeneration and necrosis of haemopoietic tissue and tubule cells with hypertrophied nuclei in *C. striatus*. Whereas, infected kidney of *C. marulius* exhibited the development of several melano-macrophage centers, hypertrophied nuclei of tubule cells and detached epithelial cells of renal tubules. Therefore an attempt has been made to record the first naturally occurring pathogenicity of *E. heterostomum* in kidney of freshwater murels.

Keywords *Euclinostomum heretostomum* · Pathology · Murels · Kidney · Histology

Introduction

Freshwater murels of the family Ophiocephalidae harbour a good number of helminth parasites. Metacercarial infections particularly those of *Euclinostomum heterostomum* show high prevalence in these fish. Usually digenetic metacercariae which occur in the liver and other visceral organs cause severe melanosis of the liver and visceral fibrosis

(Mitchell et al. 1983). Metacercaria of *E. heterostomum* have been studied by Bhalerao (1942), Srivastava (1950), Jaiswal (1957), Gupta and Agarwal (1983), Jhansilakshmibai and Madhavi (1997). Metacercariae inhabits every major organs of the host body like liver, heart, gills, body cavity etc. and cause severe pathologies to the body tissues (Rai and Pande 1969; Orecka-Grabda 1991; Laxma Reddy et al. 2006).

Kidneys are not observed to be the main target organs of parasitic invasion in many freshwater fishes. Some of the recent histopathological investigations carried out on liver are done by Harris et al. (2005), Onuoha (2010), Kaur et al. (2012). Therefore, an attempt has been made to record the histopathological changes induced by *E. heterostomum* in the kidneys of the host fishes *Channa striatus* and *C. marulius*.

Materials and methods

Collection of fish specimens

For purpose of present investigation, *C. striatus* and *C. marulius* were chosen since it is a hardy, edible and amply available fish at Bhopal. Freshly died specimens of host fishes were collected from the Lower Lake and also from the local fish markets of Bhopal. They were brought to the laboratory and examined morphologically and internally for the occurrence of helminth parasites. The host fish, *C. striatus* and *C. marulius* were collected continuously for 1 year at regular intervals.

Collection of parasites

Fish specimens were dissected out in physiological saline (0.75 % NaCl solution) for collecting helminth parasites.

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Encysted trematodes were collected from the kidneys. Collected parasites were fixed in hot alcohol-formol-acetate (AFA) solution, dehydrated and stained with acetocarmine to prepare permanent slides. The parasites were identified according to the keys given by Yamaguti (1958), Gibson et al. (2001) and Jhansilakshmibai and Madhavi (1997).

Histo-pathological study

The infected kidneys of *C. striatus* and *C. marulius* were taken out and fixed in alcoholic Bouin's fluid for 24 h. After complete removal of picric acid, the tissue was dehydrated in ethanol series (30, 50, 70, 90 and 100 %), cleared in xylene and processed for preparation of paraffin wax blocks. The tissue was then cut at 4–5 μ m thickness by rotary microtome and stained routinely with haematoxylin and eosin (H–E) for histo-pathological examination (Luna 1968). Stained histo-pathological sections were examined under Olympus research microscope. Histo-pathological changes observed were photographed and interpreted in comparison to the work of others.

Results

Out of 150 specimens of *C. striatus* and 100 specimens of *C. marulius* examined, 72 and 54 were found infected with *Euclinostomum* metacercariae, respectively. They showed the maximum prevalence of 48.0 % (± 5.2) and 54.0 % (± 1.3) in *C. striatus* and *C. marulius*, respectively. It is found encapsulated in the body cavity, muscles, liver, gill opening, intestine, kidneys and ovaries of host fishes. According to its occurrence, the body cavity and liver is most favorable site of infection whereas least infection was found in ovaries.

Morphologically, the kidney of fishes is a compact structure, the bulk of which is composed of haemopoietic or lymphoid tissue. During present investigation, a numbers of (average 2–4) deep yellowish colored encysted metacercariae were observed on the external surface of kidney, which is also partially embedded in the tissue of (Fig. 1). In a cross section of uninfected kidney, the uriniferous tubules are seen composed of prominent cells with centrally placed nuclei. The lumen of the tubule is spacious and circular. The glomeruli appear in the form of well built bunch of blood capillaries in sound condition. Scattered here and there may be seen numerous blood capillaries in well defined lymphoid tissue (Fig. 2).

The histo-pathological investigation of *E. heterostomum* infected kidney of *C. striatus* exhibited the presence of encysted metacercaria into its tissue. As, maximum pathological effect was observed in kidneys of *C. striatus*

Fig. 1 Photograph showing the kidney (K) of *C. marulius* infected by encysted metacercaria of *E. heterostomum* (arrows)

Fig. 2 Microphotograph of a cross section of normal kidney showing renal tubules (RT) and hematopoietic tissue (HPT) $\times 100$

because of the presence of 4–5 number of embedded metacercaria. The cyst wall was in close association with haemopoietic tissue (Fig. 3). Pathological effect included reduced size of glomeruli, severe degeneration and necrosis of haemopoietic tissue and tubule cells with hypertrophied nuclei (Figs. 4 and 5).

The histo-pathological investigation of *E. heterostomum* infected kidney of *C. marulius* exhibited the presence of encysted metacercaria with in a capsule, which may be formed of inner fibrous layer of parasitic tissue and outer fibrous layer of host tissue. The presence of encysted

Fig. 3 Microphotograph of a cross section of kidney of *C. striatus* infected by *E. heterostomum* showing parasite (P) and cyst wall (CW) $\times 20$

Fig. 4 Microphotograph of a cross section of kidney of *C. striatus* infected by *E. heterostomum* showing the association between glomeruli and cyst wall (AGCW) $\times 100$

Fig. 5 Microphotograph of a cross section of kidney of *C. striatus* infected by *E. heterostomum* showing necrosis (N), degeneration of hamopoietic tissue (DHPT), reduced size of glomeruli (RSG) and hypertrophied nuclei (HN) $\times 400$

Fig. 6 Microphotograph of a cross section of kidney of *C. marulius* infected by *E. heterostomum* showing parasite (P), association between glomeruli and cyst wall (AGCW) $\times 20$

metacercariae resulted in compression of host tissue (Fig. 6). Pathological effects resulted in the loosening of haemopoietic tissue and development of several melano-macrophage centers. Hypertrophied nuclei of tubule cells, detached epithelial cells of renal tubules and occlusion of tubular lumen may also be seen (Figs. 7, 8, 9).

Fig. 7 Microphotograph of a cross section of kidney of *C. marulius* infected by *E. heterostomum* showing parasite (P), outer fibrous layer of host tissue (OFLHT) and inner fibrous layer of parasite tissue (IFLPT) $\times 40$

Fig. 8 Microphotograph of a cross section of kidney of *C. marulius* infected by *E. heterostomum* showing loosening of haemopoietic tissue (LHPT) and melano-macrophage centre (MMC) $\times 100$

Fig. 9 Microphotograph of a cross section of kidney of *C. marulius* by infected *E. heterostomum* showing detached epithelial cells of renal tubules (DECRT), occlusion of tubular lumen (OTL), hypertrophied nuclei (HN) and necrosis of uriniferous tubules (NUT) $\times 400$

Discussion

Under present study, it is observed that infected kidney of *C. striatus* and *C. marulius* exhibited severe degeneration of hematopoietic (interstitial) tissue, necrosis of uriniferous tubules and hypertrophied nuclei of renal tubules. Byrne et al. (1998) also reported more or less similar finding in Atlantic salmon in which they observed distinctive acute tubular epithelial necrosis associated with interstitial haemorrhages. Rubela et al. (2008) studied the histopathological manifestation in the kidney of *C. batrachus* experimentally infected by *Procamallanus* revealed congested blood vessels, proliferated glomeruli, rupture of Bowman's capsule wall, degenerative changes, edema, necrosis, pyknosis, karyorrhexis and karyolysis in proximal and distal convoluted tubules and cloudy swelling after 15, 30, 45 and 60 days of post-infection. They also stated a pronounced destruction in proximal convoluted tubules and glomeruli after 60 days. Under present study severe damage was observed on renal cells, which may be due to relatively secretory or excretory activity of parasite which exerts effect on renal cells.

Adang et al. (2010) experimentally studied the histopathology effect of *Ascaridia galli* infection on the liver, lungs, intestines, heart and kidneys of domestic pigeons in Zaria, Nigeria. They revealed that infected kidneys had renal tubular necrosis infiltrated with few mononuclear and polymorphonuclear cells. According to Serra-Freire (1995) Nephritis caused by immune complex in kidneys of chickens infected by *Plasmodium gallinaceum* showed granular deposits in the glomerulus, associating glomerulonephritis with acute avian malaria. Whereas Jones et al. (1996) explained membranoproliferative glomerulonephritis was found in cattle naturally infected by *Tripanosoma congolensis*. Giesecker et al. (2007) experimentally explained the transmission and pathology changes caused by *Acolpenteron ureterocetus* in the kidneys of largemouth bass *Micropterus salmoides* which includes damaged renal collecting ducts with hyperplastic epithelium and varying degrees of fibrosis. Though, under present case of natural infection more or less similar findings were observed. As *E. heterostomum* is not a kidney dominating parasite but, during study it was observed to exert distinguishable pathological changes in host fishes.

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