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Histochemical studies on the distribution of Glycogen, Protein and Lipid in Cestode parasite, *Gangesia (Gangesia) ramkaei (ramkaei)*[25]

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ABSTRACT

The present communication deals with the histochemical studies on the distribution of Glycogen, Protein and Lipid in cestode parasite, *Gangesi (Gangesia) ramkaei*. The worms were collected from freshwater catfish, *Wallago attu.* It was observed that histochemically distributed amount of glycogen, protein and lipid were large in parenchyma, reproductive organs, vitelline follicles and longitudinal muscle of tegument, but the concentration differed with the different regions of proglottids. From the observation it is clear that the worm could acquire the glycogen, protein and lipid from the host.

Figures : 09 References : 31 Table : 00
KEY WORDS : Cestode, *Gangesi (Gangesia) ramkaei*, Glycogen, Lipid, Protein, *Wallago attu*

Introduction

Histochemistry is quite often defined as the aspect of histology concerned with the identification of chemical components in cells and tissues, thereby enlightening their functional anatomy. These studies offer the opportunity to correlate the structures with functions of different organs and tissues, so broadening the area of investigation. A great deal of chemical studies have been carried out in different parasitic helminths demonstrating carbohydrates, proteins, lipids and enzymes.

A few authors investigated the histochemical localization of glycogen, protein and lipid in the cestode parasites among. These were the studies on *C. laticeps* and *H. diminuta* and *H. nana.* Workers[9] found that the glycogen was primary concentrated in the medullary parenchyma of *C. laticeps* (from *A. brama*) with the highest concentration in the posterior part of the body, surrounding the sex glands, which lacked glycogen. Many workers have gone through the studies of the protein metabolism in various species of cestodes. They are L. amino acid oxidases in *H. diminuta* and some effect of change in host physiology[7] and studied on protein complexes in the cestodes *Raillietina cesticillus*[20,21], amino acids in the hydatid fluid and germinal layer of *Echinococcus*[19], urea formation and urea cycli enzymes in the cestodes *H. diminuta*[5], absorption and excretion of amino acids in the tapeworms *Anoplocephala magina*[19] and the estimation in *Phyllobothrium filiatum* has also been worked out and made comparative investigation of protein composition of *Eubothrium crassum* and the host tissues[30]. An investigator worked on serum protein in animals, which were infected, with parasitic helminths[12]. Lipid metabolism in cestodes has been worked out to only a limited extent and most studies have confirmed quantitative and qualitative examinations of lipid content and its distribution in the tissues[31]. The role of lipid in cestode metabolism is not clear. There is no evidence that lipid acts as energy reserves in the cestodes, as they do in nematodes. The synthesis of lipid has been studied in *Hymenolepis diminuta*[10,11]. In this species only a limited capacity for fatty acid biosynthesis has been demonstrated and most of its fatty acids appear to be derived from the host.

The glycogen, proteins and lipids though have their existence in cestodes but it has not been studied extensively.

Materials and Methods

Experimental studies of the histochemistry of *Gangesia* (*Gangesia*) *ramkaei* were collected from the intestine of freshwater catfish *Wallago attu* (Bleeker) at Parbhani, M.S. India. The collected worms were fixed in specific fixatives, processed and paraffin sections of 5 to 7 ¼m thickness were cut and treated for various histochemical tests.

For glycogen, worms were fixed in Bouin's fluid. The paraffin sections were dewaxed, rehydrated and stained for general carbohydrates following Periodic Acid Schiff's (PAS) technique[23]. Some sections were counterstained with iron-haematoxylin. Best's Carmine staining method was applied for the localization of glycogen. Control sections earlier digested with filtered human saliva at 37ºC for three hours (or with alpha amylase for 20 minutes) were similarly stained, and

mounted in canada balsam. For proteins, worms were fixed in Carnoy's fluid or 10% neutral buffered formalin. For the detection of general proteins, Mercury-Bromophenol blue method[22] was employed. Millon's reaction was applied for the localization of tyrosine containing proteins. As the control, sections treated in 0.5% trypsin for about one hour at 37ºC were used. For lipids worms fixed in 10% neutral buffered formalin were processed and paraffin sections were stained with Sudan Black B and mounted in glycerin[16]. Sections treated with chloroform-methanol in the ratio 1:3 for 24 hours served as control.

Results

The histochemical localization of glycogen, protein and lipid in *Gangesia* (*Gangesia*) *ramkaei* was observed under microscope. The result showed that the worm contained moderate concentration of the glycogen in its body. The glycogen content was distributed throughout body; in scolex (Fig. 1) glycogen concentration was high in the tegument, muscles of suckers and rostellum. Immature proglottid (Fig. 2) had high in the longitudinal and circular muscles of the tegument, moderate in medullary parenchyma. In mature segments, glycogen content is distributed throughout the proglottid (Fig. 3) specially longitudinal and circular muscles of the tegumental region, testes, ovary and muscles of cirrus pouch. In gravid proglottid (Fig. 3), the concentration of glycogen was higher at the tegumental region and egg capsules. There was slight trace of glycogen content in the medullary parenchyma. Thus it could be concluded that, the worm had sufficient amount of glycogen reserve in its body tissue, for its metabolic activities.

The longitudinal sections of the worm, when observed under microscope revealed that, proteins were darkly stained in scolex, immature and mature proglottid of the worms. In the scolex region (Fig. 4), the protein content was distributed throughout the body tissue. The concentration was high in tegument, muscles, in the immature proglottid the protein was in high concentration; the muscles layer of the tegument and corticular parenchyma showed higher protein content. In the mature proglottid (Fig. 5), the protein was distributed throughout the proglottid. The concentration was high in ovary, testes, in tegumental region, lateral cortical parenchyma and longitudinal nerve cords; moderate amount of protein was present in medullary parenchyma. In the gravid proglottid (Fig. 6), protein concentration was high in testes, ovary longitudinal nerve cords, lateral cortical parenchyma and moderate amount of protein in medullary parenchyma. Thus it could be concluded that, the worm *Gangesia* (*Gangesia*) *ramkaei* had relatively high content of protein, which is utilized for their metabolic activities.

Lipid content in the scolex (Fig. 7) were distributed throughout region, the concentration was higher in the muscles of suckers, tegument and cortical region. In the mature proglottid (Fig. 8), lipid content was distributed throughout the proglottid. The lipid concentration was high in tegumental region, cortical parenchyma and in gonads, moderate in the medullary parenchyma; the lipid content was high in the proglottid as compared with anterior region. The gravid (Fig. 9) proglottid stained dark black, indicated that, the lipid content was higher than mature segments. The concentration was higher in tegumental region, more amounts of lipids were found in testes, longitudinal nerve cords, eggs, and slight traces of lipid content was present in the medullary parenchyma. Thus, it can be concluded that, the lipid content in worm had a high concentration of lipid content for the metabolic activity of the worm.

Discussion

The present findings are in general agreement with the histochemical observations made by various researchers in different cestode species.

Histochemical studies on the distribution of glycogen in *Hymenolepis diminuta* have showed that glycogen is stored in the medullary parenchyma[29]. The heaviest deposition was present adjacent to the reproductive organ, especially the ovary. In present study, the glycogen content in moderate concentration of glycogen was distributed in the body of worm, especially the high content of glycogen in scolex, suckers and rostellum, longitudinal circular muscle of the tegumental region, medullary parenchyma, testes and ovary. Thus the considerable amount of glycogen reserve was in its body tissue.

The protein metabolism of the parasites of cold blooded animals has not been studied so far, but some details are known about the parasites of mammals[17,18] Mammalian tapeworms contain considerable quantities of protein, predominantly scleroprotein. It is probable that proteins play a role of some importance in the energy production of parasites[2]. In the present study protein content was high in *Gangesia* (*Gangesia*) *ramkaei* n.sp. The high amount of protein distributed in worm of body because it is naturally available from the host tissue as there was no media to acquire proteins in parasites; these proteins are naturally available from the host tissue. These worms utilize different degree of protein for producing energy.

It was observed that the eggs isolated from the terminal proglottids contained only moderate amounts of lipids[8]. The parenchyma therefore must have contained lipids well in excess of the 31% mentioned. The same type of gradient had been observed by histochemical

Glycogen content in *Gangesia (Gangesia) ramkaei* n.sp.

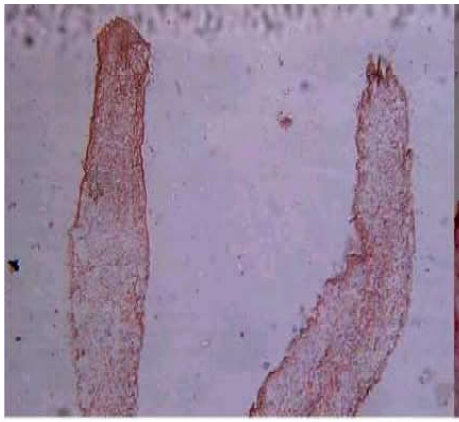

Fig. 1 : L.S. of soolex showing distribution of glycogen

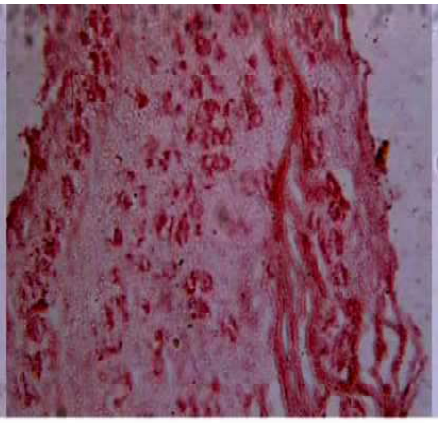

Fig. 2 : L.S. of mature proglottid showing distribution of glycogen in different parts

Fig. 3 : L.S. of gravid proglottid showing distribution of glycogen

Protein content in *Gangesia (Gangesia) ramkaei* n.sp.

Fig. 4 : L.S. of soolex showing distribution of protein

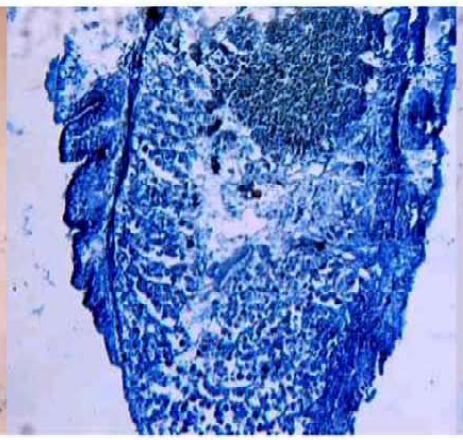

Fig. 5 : L.S. of mature proglottid showing distribution of protein in different parts

Fig. 6 : L.S. of gravid proglottid showing distribution of protein

Lipid content in *Gangesia (Gangesia) ramkaei* n.sp.

Fig. 7 : L.S. of soolex showing distribution of lipid

Fig. 8 : L.S. of mature proglottid showing distribution of lipid in different parts

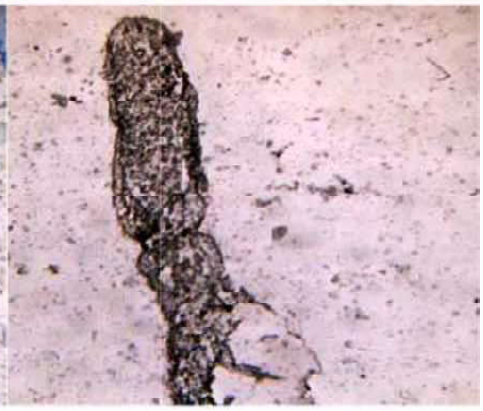

Fig. 6 : L.S. of gravid proglottid showing distribution of lipid

methods in *Hymenolepis diminuta* and in *Raillietina cesticillus*[13]. Morphologically, the most important lipid storage organ of tapeworms generally is the parenchyma. Most other organs contain either no or only little lipid. The cuticle is often lipid free, but a positive reaction was observed in the case of *Taenia taeniaeformis*. Lipid droplets are often seen in the eggs. They are usually located between the embryo and the eggshell. In some instance, e.g. *Dipylidium caninum*, large fat droplets were observed in the lumen of the uterus. Peculiar to tapeworms is the fact that some lipid occurs in the calcareous corpuscles and that in some species rather close connections between lipids and excretory system exist. Thus, in *Echinococcus granulosus* and *Moniezia expansa* fat droplets were found in the lumen of excretory canals, and in *Hymenolepis diminuta*[13] rather fat accumulations were observed around the canals. Furthermore, details concerning lipid distribution in cestodes bodies was observed by various authors[14,15,26]. In the present study sufficient amount of lipid content were distributed in the body of worm. It is essential for the normal growth. The concentrations of lipid were found in the reproductive system, tegument, cortical and medullary parenchyma, longitudinal nerve cords and neck region in both the cestodes.

Conclusion

From the histochemical observations, it was observed that the worms contain large amount of glycogen, protein and lipid in tegument at region showing deep staining reaction, parenchyma, reproductive organs vitelline follicles and longitudinal muscle of tegument. Some time the glycogen, protein and lipid content varied in different parts of the body of worm *i.e.* scolex, immature, mature and gravid proglottids. From the above observation, it is clear that the worm could acquire the glycogen, protein, and lipid from the respective host *i.e.* from the microenvironment in which they live.

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