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RESEARCH ARTICLE

HISTOCHEMICAL STUDIES ON THE DISTRIBUTION OF GLYCOGEN, PROTEIN AND LIPID IN CESTODE *GANGESIA GANGESIA RAMKAI* PAWAR, 2008

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ABSTRACT

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

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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 structure with function of different organs and tissues, so broadening the area of investigation. A great deal of chemical studies has been carried out in different parasitic helminths which demonstrating carbohydrates, proteins, lipids and enzymes. A few authors investigated the histochemical localization of glycogen, protein and lipid in the cestodes parasites among, these are the studies on *C. laticeps* was first studied by Orther Schonbach 1913), *H. diminuta* and *H. nana,* Read, Schiller and Phifer, 1958. Ginetsinskaya and Uspenskaya 1965) 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

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 by Daugherty 1955), studied on protein complexes in the cestodes *Raillietina cesticillus* by Kent 1957 a, b), amino acids in the hydatid fluid and germinal layer of *Echinococcus* by Karvavica, et al., 1959 b), urea formation and urea cycli enzymes in the cestodes *H. diminuta* by Campbell 1963 a), absorption and excretion of amino acids in the tapeworms *Anoplocephala magina* by Karvavica et al. 1959) and the estimation in *Phyllobothrium filiatum* has also been worked out, Sidorov 1980) made comparative investigation of protein composition of *Eubothrium crassum* and the host tissues. Ganzales 1978) worked on serum protein in animals, which were infected, with parasitic helminths. Lipid metabolism in cestodes has been worked out to only a limited extent and most studies have been confirmed to quantitative and qualitative examination of lipid content and its distribution in the tissues

Smyth, 1969). The role of lipid in cestode metabolism is not clear. There is no evidence for example, that lipid act as energy reserves in the cestodes, as they do in nematodes. The synthesis of lipid has been studied in *Hymenolepis diminuta* Ginger and Fairbairn, 1966 a, b). 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 its existence in cestodes but it has not been studied extensively.

MATERIALS AND METHODS

Experimental studies of the histochemistry of *Gangesia* (*Gangesia*) *ramkai* 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 µm 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 McManus Periodic Acid Schiff's PAS) technique as described by McManus 1948). 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 Mazia et al., 1953) 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 Humason, 1979) and mounted in glycerin. 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 contain moderate concentration of the glycogen in its body. The glycogen content is distributed throughout body; in scolex glycogen concentration is high in the tegument, muscles of suckers and rostellum. Immature segments also have high in the longitudinal and circular muscles of the tegument, moderate in medullary parenchyma. In mature segments, glycogen content is distributed throughout the segment specially longitudinal and circular muscles of the tegumental region, testes, ovary and muscles of cirrus pouch. In gravid segments, the concentration of glycogen is higher at the tegumental region and egg capsules. There is slight trace of glycogen content in the medullary parenchyma. Thus it can be concluded that, the worm has 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 are darkly stained in scolex, immature and mature segments of the worms. In the scolex region, the protein content is distributed throughout the body tissue. The concentration is high in tegument, muscles, in the immature segments the protein is in high concentration; the muscles layer of the tegument and corticular parenchyma shows higher protein content. In the mature segments, the protein is distributed throughout the segment. The concentration is high in ovary, testes, in tegumental region, lateral cortical parenchyma and longitudinal nerve cords; moderate amount of protein is present in medullary parenchyma. In the gravid segments protein concentration is high in testes, ovary longitudinal nerve cords, lateral cortical parenchyma and moderate amount of protein in medullary parenchyma. Thus it can be concluded that, the worm *Gangesia* (*Gangesia*) *ramkai* has relatively high content of protein, which is utilized for their metabolic activities. Lipid content in the scolex are distributed throughout region, the concentration is higher in the muscles of suckers, tegument and cortical region. In the mature segments, lipid content is distributed throughout the segment. The lipid concentration is high in tegumental region, cortical parenchyma and in gonads, moderate in the medullary parenchyma; the lipid content is high in the segments as compared with anterior region. The gravid segments stained dark black, indicate that, the lipid content is higher than mature segments. The concentration is higher in tegumental region, more amounts of lipids are found in testes, longitudinal nerve cords, eggs, and slight traces of lipid content is present in the medullary parenchyma. Thus, it can be concluded that, the lipid content in worm has a high concentration of lipid content for the metabolic activity of the worm.

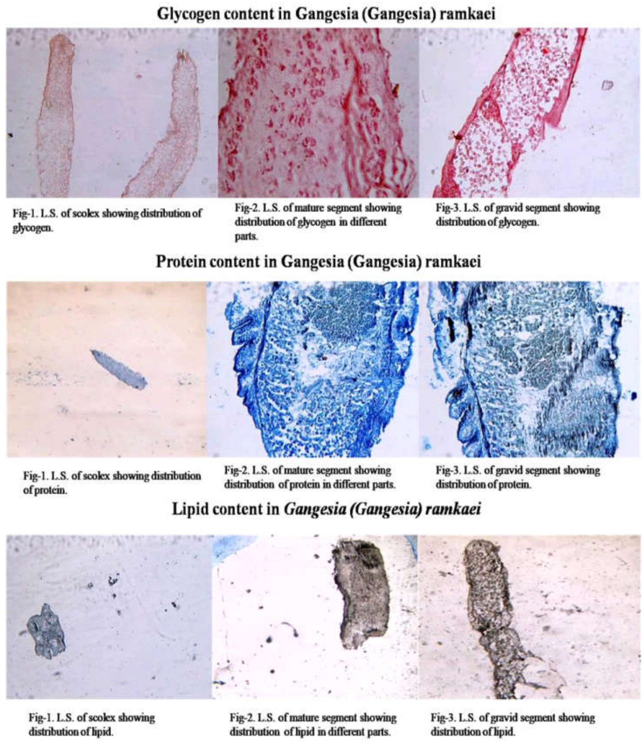

Figure. Histochemical localization of glycogen, protein and lipid in cestode *Gangesia* (*Gangesia*) *ramkai*

DISCUSSION

The present findings are in general agreement with the histochemical observations made by various researchers in different cestode species. Histochemical studies by Robert, L.S., Bueding, E. and Orell, S.A. 1970) on the distribution of glycogen in *Hymenolepis diminuta* have showed that glycogen is stored in the medullary parenchyma. The heaviest deposition is presented adjacent to the reproductive organ, especially the ovary. In present study the glycogen content is moderate concentration of glycogen are 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 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 Ivanov, 1950; Kanygina, 1951) 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 Brand, 1952). In the present study that protein content is 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 is 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.

Histochemically demonstrated the lipids in cestode parasites and found that they store only the small amount of lipid. Fairbairn et al. 1961 observed that the eggs isolated from the terminal proglottids contained only moderate amounts of lipids. The parenchyma therefore must have contained lipids well in excess of the 31% mentioned. The same type of gradient had been observed by histochemical methods in *Hymenolepis diminuta* and in *Raillietina cesticillus* by Hedrick 1958). Morphologically, the most important lipid storage organ of tapeworms generally is the parenchyma. Most other organ 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* Waitz, 1963). 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 are observed in the lumen of the uterus. Peculiar to tapeworms is the fact that some lipid occurs in the calcareous corpuscles Von Brand et al., 1960; Waitz, 1963) and that in some species rather close connections between lipids and excretory system exist. Thus, in *Echinococcus granulosus* and *Moniezia expansa* Von Brand, 1933) fat droplets were found in the lumen of excretory canals, and in *Hymenolepis diminuta* Hedrick, 1958) rather fat accumulations were observed around the canals. Furthermore, details concerning lipid distribution in cestodes bodies, observed in various authors, Hiware 1998, 1999), Rao 1960). In the present study sufficient amount of lipid content are 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 is 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 varies 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 to acquire the glycogen, protein, and lipid from the respective host i.e. from the microenvironment in which they live.

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