Proc. Indian Acad. Sci. (Anim. Sci.), Vol. 94, No. 1, January 1985, pp. 21–23.

Effect of starvation on acid phosphatase activity in *Gastrothylax crumenifer*

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Abstract. Effect of starvation on acid phosphatase activity in *Gastrothylax crumenifer* showed that activity was greater in starved individuals than in well fed ones.

Keywords. Starvation; acid phosphatase; *Gastrothylax crumenifer*.

1. Introduction

Although considerable work has been done on the subject of stress very little is known on stress and its effect on parasites or parasitism. Starvation in flukes produces increase in membraneous autophagic vacuoles exhibiting hydrolytic activity as elicited in *Megalodiscus temperatus* (Bogitsh 1973), *Schistosoma mansoni* (Bogitsh 1975), *Haematoloechus medioplexus* (Davis *et al* 1969). A study was undertaken with a view to elucidating the effects of starvation on the activity of phosphatases in *Gastrothylax crumenifer*.

2. Material and methods

To detect the effects of starvation on phosphatase activity in *G. crumenifer*, live parasites were obtained from the rumen wall. They were transported to the laboratory in Hedon–Fleig medium and were washed thoroughly to clean off debris. One batch of worms was taken separately and assayed for phosphatase activity and considered for normal activity at 0 hr. The second batch was incubated in Hedon–Fleig medium with 0·5 % glucose (fed), one million units of penicillin and 2·5 g of streptomycin per litre as described by Thorpe (1967). To determine the effect of starvation, another batch of worms was incubated in Hedon–Fleig medium without glucose (starved). Ten to fifteen worms were placed in 200 ml experimental liquid in finger bowls and the temperature maintained at 37°C. Solutions were changed after 12 hr and thereafter at 24 hr intervals; the worms were observed twice daily. Dead worms were removed promptly. A worm was considered to be "fully alive" when moving spontaneously, "moribund" or "half alive" when sluggishly moving or responding only to mechanical stimulation and "dead" when no movement was evident or no response was obtained. Usually active worms were attached firmly to the sides of the glass jars.

Observations of the parasite's enzyme activity were made at 0 hr and at the end of 12 hr, and then at intervals of 24 hr, up to 120 hr (5 days), for both control (fed) and experimental (starved) batches. At the commencement of the 6th day, parasites in the non-nutrient medium were transferred to the nutrient (glucose) medium for measurement of any difference in activity. As no parasite survived after the 6th day, even in the

nutrient medium, the experiment had to be concluded at that stage. Phosphatase activity was assayed by the method of King and Armstrong as described by Varley (1967), using disodium phenyl phosphate (0·01 M) as substrate. The protein content of the supernatant was determined following the method of Lowry *et al* (1951), using human serum albumin as a standard.

3. Results

The phosphatase activity in *G. crumenifer* was estimated at 0 hr *i.e.*, prior to subjecting them to starvation for varying periods up to 120 hr. The phosphatase activity was greater in starved individuals than in the fed ones at 12, 24, 48 and 120 hr (figure 1). The phosphatase activity was greater in refed ones than in fed ones, at 144 hr.

4. Discussion

The results indicated that starvation/stress enhanced the acid phosphatase activity in *G. crumenifer*. No biochemical work exists describing the effects of starvation on

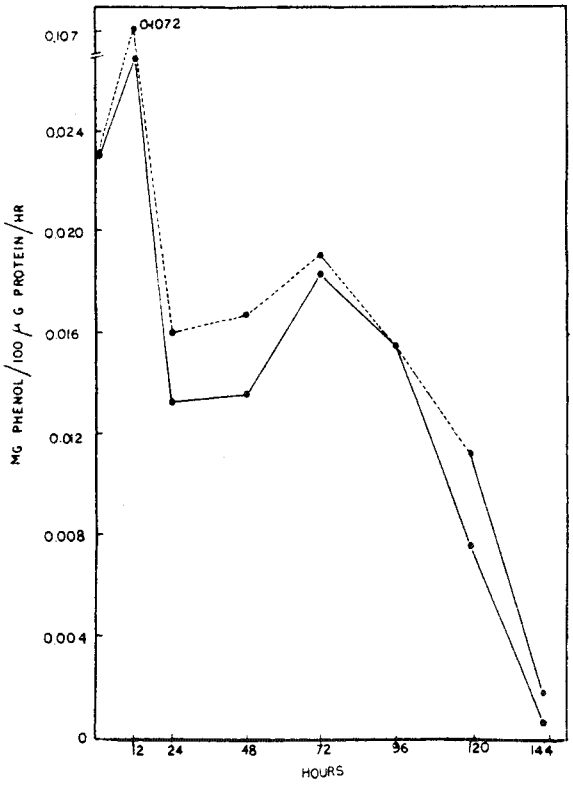

Figure 1. Phosphatase activity in *G. crumenifer* with glucose (fed) and without glucose (starved) at different hours in veronal buffer at pH 3 ((——)-fed, (– – –) starved).

phosphatase activity in digenetic trematodes. A few studies have been made histochemically on the effects of starvation on phosphatase activity in digenetic trematodes. The stimulus for the increased synthesis of phosphatases remains unknown. It is possible that the diminishing pressure of material in the lumen of the digestive tract is the triggering mechanism (Bogitsh 1975). As the amount of food in the lumen is reduced, the pressure is likewise lessened and an impulse is probably generated.

Bogtish (1973) suggested that starvation is a stimulus to which the gastrodermis of *M. temperatus* reacts by sequestered areas with the enclosed material subsequently being degraded. It has been reported that the golgi complexes become increasingly active in their relationship with the lysosome system in other types of organisms subjected to stress factors, such as starvation (Ericson 1969). Under conditions of stress (*e.g.*, starvation) organelle complexes are often found in increased numbers (Bogitsh 1973; Threadgold and Arme 1974). A marked increase in the number of acid phosphatase positive, membrane-bound vacuoles was reported in starved *M. temperatus* as compared to well fed worms (Bogitsh 1973).

The functional significance of this process lies in the possibility that it may represent a survival mechanism for the tissue following stress. The metabolism of the gastrodermis of *M. temperatus* may become reoriented so that the lytic rates become significantly greater than the synthetic rates (Bogitsh 1973).

It is desirable that a larger number of trematodes be investigated to determine how the stress would affect the various tissues, thus enabling a better understanding of this aspect of trematode physiology.

Acknowledgement

The author is indebted to Prof. D W Halton, for his valuable suggestions and for critically going through the manuscript. Thanks are due to late Dr Sita Anantaraman and Prof. M Anantaraman, Madras for their help in preparation of the manuscript.

References

Bogitsh B J 1973 Cytochemical and biochemical observations on the digestive tracts of digenetic trematodes: IX *Megalodiscus temperatus*; *Exp. Parasitol.* **32** 244–266

Bogitsh B J 1975 Cytochemistry of gastrodermal autophagy following starvation in *Schistosoma mansoni*; *J. Parasitol.* **61** 237–248

Davis D A, Bogitsh B J and Nunnally D A 1969 Cytochemical and biochemical observations on the digestive tracts of digenetic trematodes. III Non-specific esterase in *Haematoloechus medioplexus*; *Exp. Parasitol.* **24** 121–129

Ericson J L E 1969 Mechanism of cellular autophagy in *Lysosomes in biology and pathology* (eds) J T Dingle and H B Fall (Amsterdam: North Holland) Vol 2

Lowry O H, Rosenbrough N J, Farr A L and Rundall R F 1951 Protein measurement with the folin phenol reagent; *J. Biol. Chem.* **193** 265–275

Thorpe E 1967 Histochemical study with *Fasciola hepatica*; *Res. Vet. Sci.* **8** 27–36

Threadgold L T and Arme C 1974 Electron microscope studies of *Fasciola hepatica* XI Autophagy and parenchymal cell function; *Exp. Parasitol.* **35** 389–405

Varley H 1967 *Practical clinical biochemistry* 4th edn (New York: William Heinmann-Medical Books Ltd and Interscience Books Inc) p. 802