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Investigation of phosphatases in *Ascaridia galli*, A Bird Nematode parasite

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Abstract

Phosphates play an important role in metabolic activities especially in carbohydrate metabolism. The phosphatases constitute a large group of enzymes that are involved in many important phases of intermediary metabolism. They are differentiated into three types i.e. A) pyrophosphatases (inorganic and organic) B) phosphomonoesterases C) phosphodiesterases. Depending upon the substrates that are singly or doubly esterified phosphatates to which these are specific, respectively. These group, in turn, has two sub groups one of low and the other of high specificity. The phosphomonoesterases of low specificity will have optimum activity either in the acid or alkaline range, depending upon which these can be acid or alkaline phosphatases which is recovered from Ascaridia galli Schrank [5] from Nanded region (M.S.) India.

Key Words: Phosphatases, *Ascaridia galli,* Nanded region

Introduction

Phosphomonoesterases are important enzymes, responsible for the cleavage of the sugar phosphates and play an important role in metabolic processes. These were differentiated in to acid and alkaline phosphatases in present investigation. The term acid and alkaline phosphatases are used to describe the enzyme activity as a function of pH. However, the acid and alkaline phosphatases are group names and do not indicate well defined entities, Von Brand [6].

The study of phosphatases are helpful to understanding the rate of sugar transport in the tissues of the parasite and its uptake from the intestinal contents of the host. Phosphatases exhibit varying properties from species to species and also between different developmental stages of particular species. Various investigators have described the role of phosphates in the parasitic helminthes. The cuticle possesses microvilli or microtriches which are metabolically very active. These structures resemble the brush border of invertebrate cells. These cells contain hydrolytic enzymes and are involved in digestion and absorption function [1].

The non specific monoesterases of *Ascaridia galli* have been performed by Pavlov and Chesnokava [4]. As there is no information available about the acid and alkaline phosphataese activity levels in male and female nematode parasites of *Ascaridia galli* infecting naturally domestic fowl. (Gall*us gallus domesticus*).

For understanding the metabolic differentiation the activity levels of acid phosphatase (pH 3.5 to 6.5) and alkaline phosphatase (pH 7.5 to 10.5) were investigated.

Materials and Methods

The activity of acid phosphatase (orthophosphorlc monoester phosphohydrolase, EC 3.1.3.2) and alkaline - phosphatase (orthophosphorlc monoester phosphohydrolase, EC 3.1.3.1) was determined by estimating the Pi liberated by the method of Fiske and Subba Row [2].

10% homogenates of male and female worms were prepared in distilled water. The supernatant, obtained, after centri¬fugation at 2500 rpm for 15 minutes, was used as crude enzyme extract. The assay mixture contained 4.5 ml of acetate buffer and 0.5 ml of enzyme extract (equivalent to 50 mg tissue). The buffer was taken at different pH ranges, viz: 3.5, 4.5, 5.5, 6.5, 7.5, 8.5, 9.5 and 10.5 for the estimation of phosphatase activities in both acid and alkali ranges. All the tubes were incubated at 37°C for an hour. After incubation the reaction was stopped by adding one ml of 30% TCA. Zero hour controls were maintained with one ml of 30% TCA before the addition of enzyme extract. The samples and zero time controls were centrifuged at 2000 rpm for 15 minutes. To 1 ml of this supernatant, in each case, 0.5 ml of 2.5% (w/v) ammonium molybdate and 0.2 ml of ANS reagent were added. After dilut¬ing the samples, controls and blank to 10 ml, the colour developed was read at 660 m . The enzyme activity was expressed in terms of Pi liberated/hour/100 mg tissue.

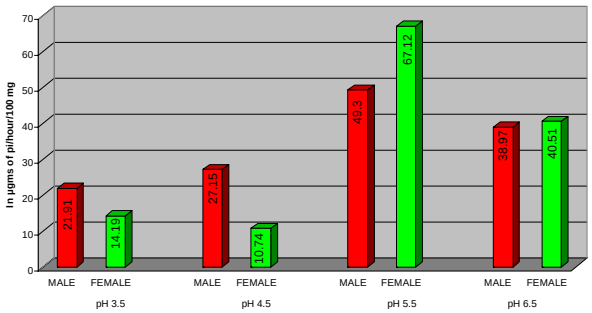
Result and Disscution

Acid phosphatase activity in male and female nematode parasites of *Ascaridia galli*.

Sr. No.	Parameter	Activity Male (5) MEAN ± S.D.	Female (5) MEAN ± S.D.	Male to Female ratio	Percentage difference (%)
1	pH 3.5	21.91 ± 1.97	14.19 ± 0.49		
2	pH 4.5	27.15 ± 2.88	10.74 ± 0.57		
3	pH 5.5	49.30 ± 6.12	67.12 ± 3.92	0.68	22.43
4	pH 6.5	38.97 ± 6.10	40.51 ± 1.95		

- Values puted in parentheses showing number of samples estimated.
- Values expressed In µgm of pi/hour/100 mg.

ACID PHOSPHATASE ACTIVITY IN MALE AND FEMALE NEMATODE PARASITES OF *ASCARIDIA GALLI*.

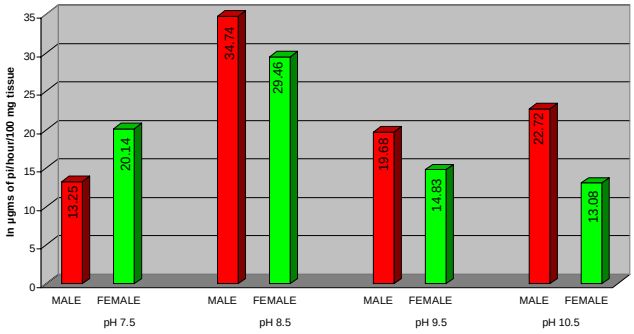


Alkaline phosphatase activity in male and female nematode parasites of A*scaridia galli*.

Sr. No.	Parameter	Activity Male (5) MEAN ± S.D.	Female (5) MEAN ± S.D.	Male to Female ratio	Percentage difference (%)
1	pH 7.5	13.25 ± 2.35	20.14 ± 2.67		
2	pH 8.5	34.74 ± 4.08	29.46 ± 3.97		
3	pH 9.5	19.68 ± 3.22	14.83 ± 1.92	1.54	17.48
4	pH 10.5	22.72 ± 2.20	13.08 ± 2.14		

- Values puted in parentheses showing number of samples estimated.
- Values expressed in µgm of pi/hour/100 mg tissue.

ALKALINE PHOSPHATASE ACTIVITY IN MALE AND FEMALE NEMATODE PARASITES OF *ASCARIDIA GALLI*.



The values obtained from activity levels are tabulated in observation table for acid phosphatases and for alkaline phosphatases, same are represented in graph. From statistical data received we can state that the acid phosphatases activity is maximum at pH 5.5 and the alkaline phosphate activity is maximum at pH 8.5 in both the sexes of nematode parasite. The optimum values for acid phosphatase activity in females is 67.12 ± 3.92 μgm pi/hour/100 mg wet weight and in males is 49.30 ± 6.12 μgm pi/hour/100 mg wet weight. The activity ratio of male to female is 0.68 μgm pi/hour/100 mg wet weight suggesting that its activity is about 22% less in males.

The optimum alkaline phosphatase activity in male is 34.74 ± 4.08μgm pi/hour/100 mg wet weight, and in female is 29.46 ± 3.97 of μgm pi/hour/100 mg wet weight, respectively. Male to female ratio is 1.54 μgm pi/hour/100 mg wet weight and the difference is 17.48% It state that male consume more alkaline phosphatases, than female nematode parasites.

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