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Research Article

## Changes in Acid Phosphatase and Alkaline Phosphatase activity in gonads of Rainbow trout

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**Abstract:** The present study describes the activity of acid phosphatase and alkaline phosphatase in the ovaries and testis of rainbow trout during different phases of reproduction. The activity of acid phosphatase as well as alkaline phosphatase was found to be maximum in mature stage and was minimum in spent stage in rainbow trout ovary. However in the testis the activity of acid phosphatase was observed maximum in spent stage and minimum in maturing stage. The activity of alkaline phosphatase activity was observed maximum in mature stage and was minimum in immature stage in trout testis.

**Keywords:** Rainbow trout, Acid phosphatase, Alkaline phosphatase, Immature, Maturing, Mature and Spent stage.

### INTRODUCTION

Specific biochemical changes occur during the development and growth of the fish gonads. These metabolic activities are controlled by different enzymes. Phosphatase is a hydrolytic enzyme, leading to the release of ortho-phosphate from phosphorus compound and based on the optimum pH of action environment, classified into acid phosphatase (ACP, EC 3.1.3.2, optimum  $\text{pH} \leq 6.0$ ) and alkaline phosphatase (ALP, EC 3.1.3.1, optimum  $\text{pH} \geq 8.0$ )<sup>1</sup>. Acid phosphatase is a widely distributed enzyme it releases inorganic phosphate, which is regarded a key metabolite for cellular development<sup>2</sup>, alkaline phosphatase is an intrinsic plasma membrane enzyme and is found in almost all animal cells<sup>3</sup>. Both Acid Phosphatase as well as Alkaline Phosphatase is involved in various metabolic processes such as permeability, growth and cell differentiation, protein synthesis, absorption and transport of nutrients, for gonadal maturation<sup>4</sup>. The alkaline phosphatase plays an important role by providing inorganic phosphate

(Pi) to the tissue. The inorganic phosphate is required in the synthesis of several metabolites during developmental stages, change in the acid and alkaline phosphatase activities affects in the metabolism of the fish. Phosphatase activities change has been regarded as indices of growth, illness and spawning of fish<sup>5,6</sup>. Shaffi *et al*<sup>7</sup>, have recorded maximum alkaline phosphatase activity during the maturation stage of *Clarias batrachus*.

Studies on the Enzyme assay particularly Acid Phosphatase and Alkaline Phosphatase changes during the development of gonads in rainbow trout are still fragmentary. Keeping this lacunae in view, the present work has been proposed to analyse changes in (acid phosphatase and alkaline phosphatase) contents of the ovaries and testis of rainbow trout of Kashmir waters during different stages of development.

## MATERIALS AND METHODS

On the basis of detailed seasonal morpho- histological changes in the gonads of Rainbow trout, the following stages of development viz. the immature, maturing, mature and spent have been studied. The important morpho-histological changes have also been observed during immature, maturing, mature and spent stages, therefore, the present biochemical study has been conducted during these stages of maturity. Sexually matured fishes were captured from Kokernag (33.69° N and 75.22° E) and Verinag (33.55° N and 75.25° E). For the determination of phosphatases: Acid phosphatase and Alkaline phosphatase activity were measured by method of Linhardt and Walter<sup>8</sup>. Enzyme assay included 0.2 ml homogenate and 0.8 ml of citrate buffer (pH 4.0) containing  $5.5 \times 10^{-3}$  M p- nitrophenyl phosphate. Mixture was incubated at 40 °C for 30 min. Reaction was stopped by addition of 4 ml of 0.1 N NaOH after incubation. The absorbance was measured at 405 nm on spectrophotometer.

**Calculations :** Acid phosphatase and alkaline phosphatase activity is calculated by the following formula

$$\text{Units/ml of sample} = (\text{DA}_{410} \text{ Total}) \times (1.9)/(2) \times (18.3) \times (0.6)$$

Where:

1.9= Total volume of reaction in ml

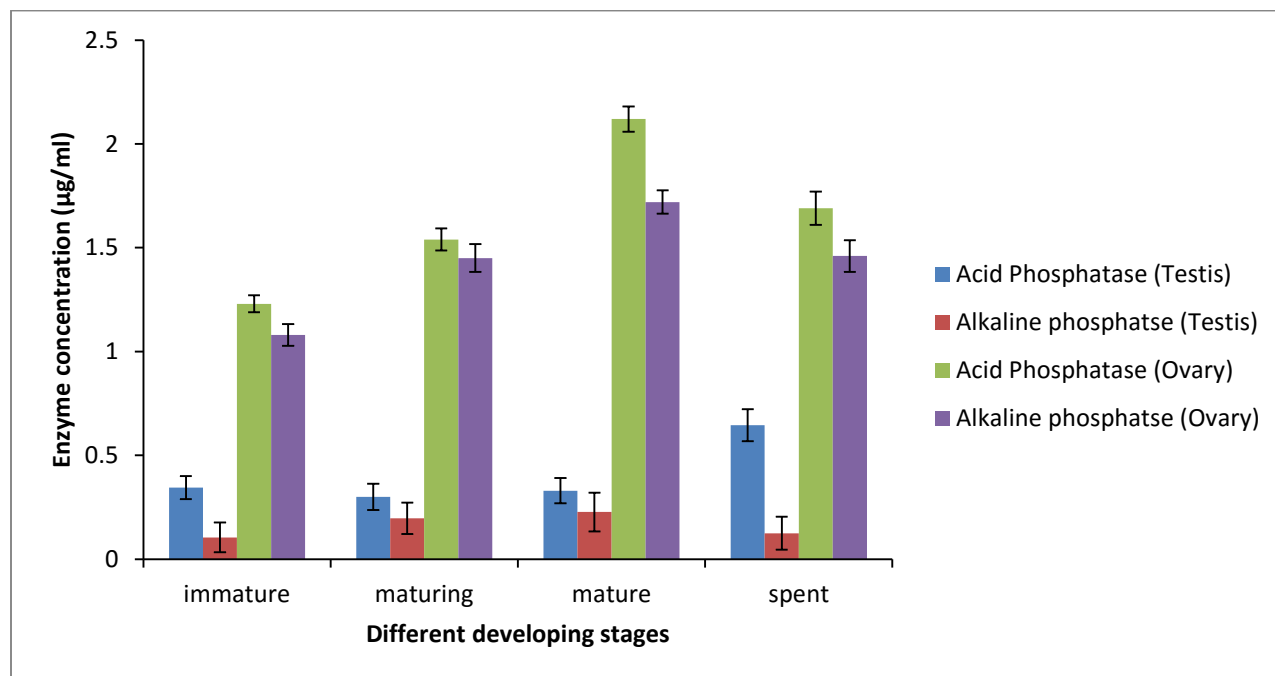
2= Time of reaction in minutes

18.3 = Millimolar absorptivity (extinction coefficient) of p-nitrophenol at 405 nm at alkaline pH

0.1 = Initial volume of serum or alkaline phosphatase sample used in ml

## RESULTS

In the ovaries of rainbow trout the activity of acid phosphatase begins to increase during maturing stage and its activity was found to be maximum in matured stage and then decreases in spent stage. The activity of the alkaline phosphatase showed a similar pattern in the ovary of rainbow trout (**Table 1, Fig 1**). Acid phosphatase activity was found to be maximum in spent stage and minimum in mature stage in trout testis. However the activity of alkaline phosphatase activity begins to increase from maturing stage and was highest during matured stage (**Table 1, Fig 1**).



**Fig. 1:** Acid and alkaline phosphatase activity in rainbow trout testis and ovary during different phases of development

**Table 1:** Seasonal variations in acid phosphatase and alkaline phosphatase activity in the ovary and testis of rainbow trout

Stages	Acid phosphatase (ovary)	Alkaline phosphatase (ovary)	Acid phosphatase (testis)	Alkaline phosphatase (testis)
Immature (stage I)	1.23	1.08	0.345	0.105
Maturing (stage II)	1.54	1.45	0.30	0.197
Mature (stage III)	2.12	1.72	0.33	0.227
Spent (stage IV)	1.69	1.46	0.645	0.125

## DISCUSSION

The alkaline phosphatase activity in the ovaries of Rainbow trout was found to be maximum in matured stage and the minimum activity was recorded in immature stage, from the immature stage to matured stage initially the activity begins to increase and thereafter the activity of alkaline phosphatase begins to

fall in spent stage. Similar types of variations were also recorded in the ovaries of *Clarias batrachus* during different stages of breeding cycle<sup>7</sup>. The alkaline phosphatase activity increased in the ovaries of *Puntius chilinoides* during the maturing stage, increase in alkaline phosphatase during maturing stage indicates that synthesis of new proteins takes place as alkaline phosphatase is found to be involved in protein synthesis. The acid phosphatase activity in rainbow trout ovary was maximum in matured stage and thereafter it begins to decrease in spent stage. The acid phosphatase activity was recorded minimum in immature stage. In the present study acid phosphatase activity in testis was found to be maximum in spent stage and minimum in maturing stage. It is believed that acid phosphatase plays an important role in fish ovaries by synthesizing essential metabolites by liberating inorganic phosphate. In case of fish testis activity of acid phosphatase reveals that it may play fewer roles during spermatogenesis. The activity of alkaline phosphatase was found to be maximum in mature stage and lowest in immature stage, after mature stage the level of alkaline phosphatase begins to fall up to immature stage, indicating that the alkaline phosphatase plays an important role in the development of sperms. Nauriyal and Singh<sup>9</sup> have observed that in testis of *Puntius chilinoides* the activity of acid phosphatase was minimum during maturing stage and was highest at the immature stage. The alkaline phosphatase activity was found to be lowest at maturing stage and thereafter it increases up to spent stage.

## CONCLUSION

The present results supplied valuable information of the seasonal variation activity of Acid phosphatase and Alkaline phosphatase. The study reveals that Alkaline phosphatase plays an important role in protein synthesis and development of sperms. While acid phosphatase is believed to play an important role in fish ovaries by synthesizing essential metabolites.

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