between the morula and yolk plug stage was the difference not significant. Further, within these 3 advanced stages *t* was not significant at 5% level. In other words, yolk plug stage is the earliest which could be selected for determining the percentage of fertilization and estimates of hatchlings showing minimum variation. From the biological point of view, it was found that irregular cell division and whitening of eggs are more frequent in the earlier stages and once the yolk plug stage is attained, the probability of hatching is high. The yolk plug stage may be reasonably selected for qualitative assessments considering the practical necessity to assess at the earliest.

The matrix diagram (figure 1) is a new approach in presenting the results of pairwise comparisons most effectively. Using distinct patterns, significant differences at different levels can also be presented compactly. Inferences can be drawn more efficiently. One half of the diagonal can be used to present *t* values or other statistics used, if required. Another advantage is that the graph giving related statistics, in this case, mean standard deviations and CV can also be incorporated in the matrix diagram, rendering biological interpretation easier.

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NEUROENDOCRINE INVOLVEMENT IN THE REGULATION OF LACTATE AND SUCCINATE DEHYDROGENASE ACTIVITIES IN THE FRESHWATER CRAB, *BARYTELPHUSA GUERINI* (H. MILNE EDWARDS) (DECAPODA, POTAMIDEA)

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THE oxidative enzymes are intimately concerned with oxygen consumption and oxidation of metabolites. The neuroendocrine control of respiratory metabolism and oxidative enzymes in crustaceans is now an accepted fact although the effects of eyestalk ablation and eyestalk extract injection vary in different animals[1–9]. Also, the probable means by which the variations in respiration are brought about by the eyestalk hormone is not worked out clearly. As such, more studies on regulation of the activities of oxidative enzymes by the eyestalk hormone are needed for understanding the mechanism of neuroendocrine regulation of respiratory metabolism.

An earlier study on the crab, *Barytelphusa guerini* recorded that the respiratory metabolism declines on eyestalk ablation and is restored to the normal levels by the injections of eyestalk extracts[5]. In view of this, alterations in lactate and succinate dehydrogenase activities in relation to eyestalk ablation and eyestalk extract injections are analysed to assess the possible mechanism of neuroendocrine involvement in the regulation of respiratory metabolism in this animal. Collection maintenance, choice of animals, bilateral eyestalk ablation and eyestalk extract injections were carried out according to the procedure described earlier[5].

Succinate dehydrogenase (E. C. 1. 3. 99. 1) and lactate dehydrogenase (E. C. 1. 1. 1. 27) activities were estimated quantitatively in the muscle, gill, heart and hepatopancreas tissues in (i) the normal animals with intact eyestalks, (ii) in the eyestalk ablated animals, and (iii) in the eyestalk ablated but eyestalk extract injected animals. Eyestalk ablated animals were maintained for 48 hr and divided into 2 batches. One batch was used for enzyme assay,

while the other batch received eyestalk extract injections and used for enzyme assay 48 hr after injections.

The enzyme assays were performed according to the method of Nachlas *et al*[10] and the enzyme activity was expressed as μmol formazon/g wet wt/hr. A minimum of 6 observations were made for each assay and the results were statistically analysed using students' *t* test keeping the level of significance $P = 0.05$

Both the SDH and LDH activities could be demonstrated in all the four tissues studied but the levels of activities were different amongst them. SDH activity decreased in the order of heart > hepatopancreas > muscle > gill, while LDH activity showed a decrease in the order of hepatopancreas > muscle > gill > heart. SDH was greater in heart, while LDH was slightly higher in hepatopancreas, but there was not much difference in the activity of two enzymes in the muscle and gill.

Bilateral extirpation of eyestalks led to a decrease in SDH activity in all the tissues. This decrease in SDH activity which was about 69.40% in muscle, 46.74% in gill, 42.97% in heart and 64.81% in hepatopancreas after 48 hr of eyestalk ablation was also highly significant ($P < 0.01$), when compared to the normal values. Injection of eyestalk extracts into eyestalk ablated animals increased SDH activity to that of the normal level by about 48 hr. The recovery of the activity was almost complete, such that the variations in SDH activity of experimental animals (eyestalk extract injections received) were insignificant ($P > 0.1$), when compared with normal intact animals and were highly significant ($P < 0.01$) in respect of the control eyestalk ablated animals (figure 1. A_1–A_4).

LDH activity increased by 60.12% in muscle, 66.18% in gill, 49.22% in heart and 52% in hepatopancreas after 48 hr of eyestalk removal and this increase in activity was also highly significant ($P < 0.01$), when compared to the normal values. Injections of eyestalk extracts into eyestalk ablated animals brings down LDH activity to the normal level by about 48 hr. The recovery in activity was almost complete such that the variations in LDH activity of experimental animals were insignificant ($P > 0.05$), when compared to the normal but were highly significant ($P < 0.01$) in comparison with control values (figure 1. B_1–B_4).

The present investigation demonstrates the occurrence of both LDH and SDH in different tissues tested suggesting thereby the operation of both glycolytic and tricarboxylic acid cycles in these

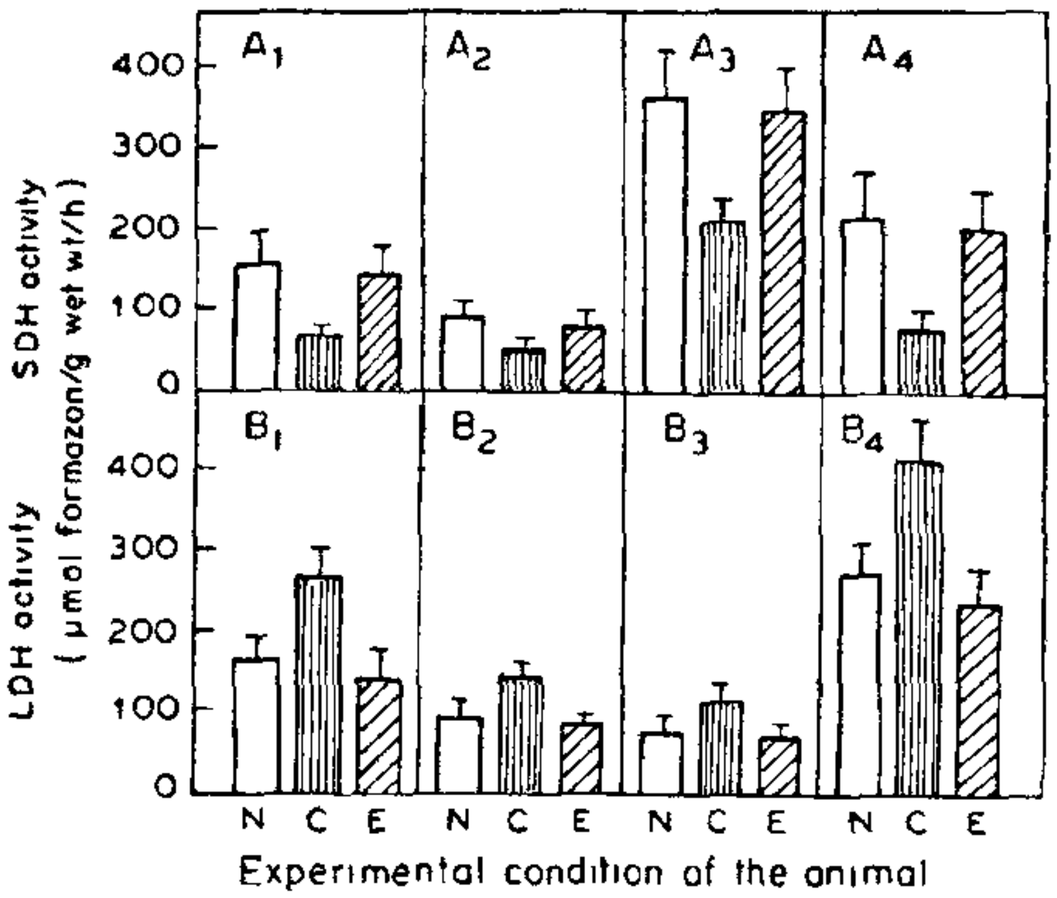

Figure 1. Succinate (A) and lactate (B) dehydrogenase activities in the muscle (1), gill (2), heart (3) and hepatopancreas (4) tissues of the normal animals with intact eyestalks (N) eyestalk ablated (C) and eyestalk ablated experimental animals receiving eyestalk extract injection (E). (The values shown are the average of 6 observations ± S. D.).

tissues. The activity levels of these two enzymes are different in the different tissues indicating that two metabolic pathways operate at different levels in these tissues. Thus the heart shows to be predominantly aerobic metabolism, whereas the hepatopancreas shows slightly greater anaerobic metabolism. However, muscle and gill are capable of anaerobic and aerobic metabolism at the same rate. While SDH activity is shown to be more in hepatopancreas than in gill in the present study, the reverse was shown to be true in *Paratelphusa hydrodromous*[7] and the activity decreases in the order gill > muscle > hepatopancreas > hypodermis[9].

Earlier studies on the neuroendocrine control of oxidative enzymes by the eyestalk hormones are not exclusive and do not bear a good conclusion with variations in oxygen consumption under similar conditions. A marked increase in the activity of SDH in hepatopancreas of *Soylla serrata*[4] and *Oziotelphusa senex senex*[9] on eyestalk ablation and its return to the normal level by eyestalk extract injection is evident — thus paralleling the trend obtained for oxygen consumption in these animals[2,8]. The increase in metabolic rate is shown to be due to the stimulation of eyestalk ablation[4,9] But similar increases in muscle and hypodermis are not influenced. Also, the activity of malate dehydrogenase and cytochrome oxidase is not affected by

eyestalk extract injection[9]. Elevation in isocitrate and glucose-6-phosphate dehydrogenase activities on eyestalk ablation is also noticed in *Oziotelphusa senex senex* especially in the hepatopancreas. This provides further evidence for the endocrine control of oxidative enzymes and for the presence of an active principle that inhibits respiratory metabolism[6].

Eyestalk extracts stimulate oxygen consumption of muscle homogenates in *Astacus laptodactylus* and this could be due to its stimulating effect on TCA cycle enzymes[1]. In contrast, the activities of the oxidative enzymes such as isocitrate dehydrogenase, cytochrome oxidase, SDH and malate dehydrogenase increase upon destalking of this animal[3].

The respiratory metabolism in the crab, *Barytelphusa guerini* decreases on bilateral eyestalk extirpation and rises to normal levels upon injections of eyestalk extracts[5]. As a corollary to this the present study shows that the enzymes of glycolysis and Krebs TCA cycle undergo typical variations in relation to eyestalk ablation and injection of eyestalk extracts in all the four tissues studied. This permits us to suggest a possible basis for the variations in the metabolic rate in relation to eyestalk principle. The decrease in metabolic rate on eyestalk ablation may be due to suppression of the TCA cycle and acceleration of the glycolytic pathway. Conversely, restoration of metabolic rate on eyestalk extract injection may be due to acceleration of the TCA cycle and suppression of glycolytic pathway. It is possible that the eyestalk hormone facilitates the operation of the TCA cycle by favouring the entry of pyruvic acid into the TCA cycle. Low level of LDH activity prevents conversion of pyruvic acid to lactic acid and thus the glycolytic pathway is inhibited. High SDH activity at the same time promotes the operation of the TCA cycle by channelising more pyruvic acid into the TCA cycle by the formation of acetyl-co-A and citric acid.

The loss of metabolic acceleratory factor through eyestalk ablation results in increase in the LDH activity and decrease in SDH activity as well as the general metabolic rate. Its replenishment through the injection of eyestalk extracts into eyestalk ablated animals drops LDH activity and elevates SDH activity, which leads to restoration of general metabolic rate to the normal level. Thus the control of respiratory metabolism by the eyestalk principle may be due to its stimulatory effect on the enzymes of the TCA cycle and deceleratory or inhibitory effect on those of the anaerobic glycolysis.

Although this explanation looks sound further evidence by way of demonstration of variations in cytochrome oxidase, ATP levels and lactic acid levels etc would be helpful for confirming this view. Studies along these lines are yielding encouraging results. However, data obtained by the earlier workers support the present view that the neuroendocrine control of respiratory metabolism is achieved by the regulation of the activities of oxidative enzymes.

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NEW RECORDS OF NATURAL ENEMIES OF *SPODOPTERA LITURA* (FAB.) IN KOLHAPUR, INDIA

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THE survey of natural enemies has immense value in biological control of pests. *Spodoptera litura* (Rab.)