ISSN: 0974 - 0376

The Ecoscan : Special issue, Vol. VII: 123-127: 2015
AN INTERNATIONAL QUARTERLY JOURNAL OF ENVIRONMENTAL SCIENCES
www.theecoscan.in

# CHANGES IN THE RATE OF OXYGEN CONSUMPTION OF FRESH WATER BIVALVE, *INDONAIA CAERULEUS* (PRASHAD, 1918) WITH INJECTIONS OF CEREBRAL GANGLIONIC EXTRACT AND EQUIVALENT COMMERCIAL HORMONES DURING SUMMER SEASON

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**KEYWORDS**

Cerebral ganglionic extract
Progesterone
Estradiol
Oxygen consumption
Freshwater bivalve

**Proceedings of National Conference on Harmony with Nature in Context of Bioresources and Environmental Health**
(HORMONY - 2015)
November 23 - 25, 2015, Aurangabad,
organized by
Department of Zoology,
Dr. Babasaheb Ambedkar Marathwada University
Aurangabad (Maharashtra) 431 004
in association with
National Environmentalists Association, India
www.neaindia.org

The Ecoscan
AN INTERNATIONAL QUARTERLY JOURNAL OF ENVIRONMENTAL SCIENCES

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## ABSTRACT

Considering the importance of neuro-endocrine controlon the metabolic activities in freshwater bivalves, we report here the effect of injections of cerebral ganglionic extract and equivalent commercial hormones (Progesterone & Estradiol) on respiratory metabolism of freshwater bivalve mollusc *Indonaiacaeruleus* (Prashad, 1918) from Godavari River. During summer season, the adult bivalve mollusc, *Indonaiacaeruleus* (48-53 mm shell length) were subjected to a) control (normal) (b) injection of cerebral ganglionic extract of same species to intact individuals (c) injection of equivalent commercial hormoneprogesterone to normal control and (d) injection of estradiolto normal control for 8 days. The rates of oxygen consumption in bivalves from all four groups (including control) were measured on 2nd, 5th, and 8th day. The study revealed that, the rate of oxygen consumption was significantly decreased in cerebral ganglionic extract injected group on 2nd day. The rate of oxygen consumption also showed significant decrease in progesterone injected group on 5th day and all the three groups on 8th day. The rate of oxygen consumption showed more decrease on 8th day in ganglionic extract injected as well as progesterone injected group.

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## INTRODUCTION

In general, most of the vital activities in bivalve are regulated by neuro-endocrine centers. The respiratory rate data of the animals reflects their general metabolic rate. The existence of neuro-endocrine modulation of metabolic rate will be the adaptive significance for the freshwater bivalves. Comparatively very little work was done on the endocrine regulation in bivalve molluscsand also comparatively; very less attention has been given on the role of neuroendocrine centers in respiratory metabolism. In the field of neuro-endocrinology, such neuroendocrine control on oxygen consumption has been reported for crustaceans (Nagabhushanam and Kulkarni, 1979). Hanumante *et al.* (1980) has been shown that neurohormones from pleurovisceral ganglia regulate the rate of oxygen consumption in gastropod molluscs. The role of cerebral and visceral ganglia in respiratory metabolism has been reported for estuarine clam, *Katelysia opima* (Mane *et al.,* 1990), for freshwater bivalvemollusc,*Lamellidensmarginalis*(Vedpathak and Wagh, 2009) and *Indonaia caeruleus* (Vedpathak *et al.,* 2011).

In bivalve molluscs, two types of neuro-secretory cycles occurs in cerebral ganglia (Lubet, 1970), the short term cycle (*i.e.* sudden change in temperature, pH and salinity) and long term cycle related to certain activity of reproduction and metabolism. Such neurosecretory cycles of neurosecretory cells was reported by Nagabhushanam and Mane (1973) for estuarine clam, *Katelysia opima*.

Review of literature shows very little information is known on neuro-endocrine control of oxygen consumption in freshwater bivalves. Since many features of aerobic metabolism can be studied directed by measurement of the rate of oxygen consumption by intact animals. Several reports are available on respiratory physiology of freshwater bivalves from India and abroad (Salanki and Lukacsovice, 1967, Bayne, 1976, Zs-Nagy 1974 and McMohan, 1979).

Thus, considering the paucity of information on endogenous regulation in the respiratory metabolism (because the respiration is considered as one of the important aspect for understanding the physiological adaptation of a species. In bivalve shellfishes from the Inland waters, the present study is taken on freshwater bivalve, *Indonaia caeruleus*from Godavari river.

## MATERIALS AND METHODS

The adult freshwater bivalve, *Indonaia caeruleus* (48-53 mm in shell length) were collected from Godavari River near Aurangabad, during summer season. After brought to the laboratory the shells of the bivalves were brushed and washed with water to remove the mud and fauling fungal and algal biomass and they were acclimatized for 24 hr. in laboratory conditions. No food was given to the animals during laboratory acclimatization and subsequent experimentation. After 24 hr. the animals were arranged in four groups, each group containing 20 animals in 10 lit.of aerated water. The first group of animals was served as normal control and other three groups were experimental with (i)injection of cerebral ganglionic extract to intact control; (ii) injection of equivalent progesterone to normal intact control

and (iii) injection of equivalent commercial hormone estradiol to normal control bivalves. For injection of cerebral ganglionic extract, extract was prepared in 1:1 ice cold distilled water and ethanol (*i.e.*20 ganglia in 2mL ice cold distilled water and ethanol), it was centrifuged and injected (0.2 mL extract/animal *i.e.* equivalent to 2 ganglia/animal), into the foot (muscular region). The experiment was run for 8 days. The physico-chemical characteristics of water used in experiments i. e. temperature, pH, hardness and dissolved oxygen contents of the water were determined on every two days throughout the experimental period.

The rate of oxygen consumption of individual animal from each group was determined by modified Winkler's technique (Golterman *et al.,* 1978), in a specially prepared brown coloured respiratory jar of 1 lit capacity. Four closed respiratory jars, each with an inlet and outlet. Five animals from each group were kept in continuous circulation of water inside jar, in order to open their shell valves. Once the animals were opened their valves, the flow of water was cutoff and sample of water from it, was drawn after 1hr. For determination of oxygen consumption, the; flesh of the individual animal was taken out carefully from the shell and socked on the blotting paper to remove the excess water. Blotted flesh was then weighed to obtain the wet-weight of the individual bivalve.

The oxygen consumed by each animal was then calculated and expressed as mg O2/l/h/gm wet-weight of the flesh. The mean values of four individual animals from each group were used for statistical analysis. For confirmation of results all the values were subjected to statistical analysis using 't' test (Dowdswell, 1957). Percentage differences were also calculated in experimental group.

## RESULTS

The results of the experiments were shown in Fig. 1. The physico-chemical characteristics of the water used in experiments during summer season were - Temperature (25.0C- 28.0C); pH (7.8 - 7.92); hardness in terms of bicarbonates (120 - 159.2 ppm) and dissolved oxygen content (5.75 – 6.4 mg/L/h).

The rate of oxygen consumption was significantly decreased (0.2792 ± 0.0506, 24.65%, $P < 0.01$) from ganglionic extract injected group, on 2nd day compared to control. Similarly the rate of oxygen consumption showed significant decrease (0.3092 ± 0.0958, 23.69%, $P < 0.05$) in progesterone injected group,on 5th day. On 8th day, the rate of oxygen consumption also showed significant decrease (0.1734 ± 0.02807, 24.48%, $P < 0.001$) in ganglionic extract injected group, (0.1739 ± 0.03479, 24.26%, $P < 0.001$) in equivalent progesterone hormone injected group and (0.2063 ± 0.0353, 17.27%, $P < 0.01$) in estradiol injected group.

The rate of oxygen consumption in control group was (0.3707 ±0.06833), (0.3804 ± 0.08320, 21.66%), and (0.2296 ± 0.02436, 38.79%) on 2nd, 5th and 8th day respectively. While the rate of oxygen consumption in ganglionic extract injected to intact animals group was (0.2792 ± 0.05062, 24.68%), (0.3243 ± 0.08072, 11.67%) and (01734 ± 0.02807, 24.48%) on2nd, 5th, and 8th day respectively. The rate of oxygen consumption in progesterone injected and estradiol injected were (0.1739 ± 0.03479, 24.26%) and (0.2063 ± 0.03530, 17.27%) on 8th day respectively.

## DISCUSSION

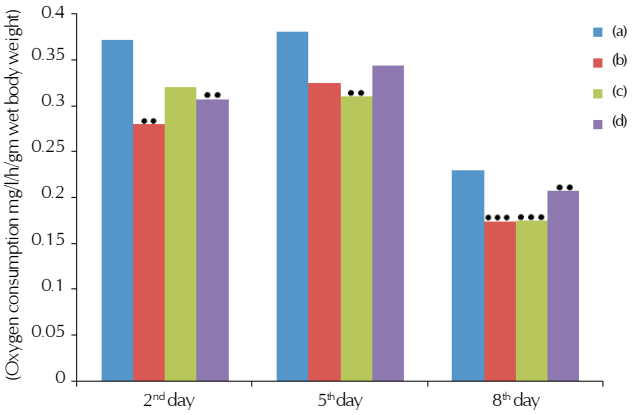
The present study on freshwater bivalve, *Indonaia caeruleus*, revealed that, injection of cerebral ganglionic extracts to intact bivalves causes significant decrease in the rate of oxygen consumption on 2nd, 5th and 8th day. The rate of the oxygen consumption in estradiol injected animals also caused significant decrease on any 2nd, 5th and 8th day compared to respective control. But the rate showed increase in all groups on 5th day compared to 2nd day.

A significant decrease in the rate of oxygen consumption after injection of ganglionic extract, progesterone and estradiol as compared to control on 2nd, 5th and 8th day suggest the possibility of feedback mechanism in regulation of oxygen consumption. The existence of possible feed back mechanism could be because of further stimulation of rate of oxygen consumption after injection of cerebral ganglionic extract to the intact animals, which is receiving the cerebral ganglionic extract and restore the rate of oxygen consumption.

From the data, it can be suggested that cerebral ganglia must possesses the hormonal factor which is responsible for regulation of oxygen consumption. Injection of cerebral ganglionic extract to the ganglia removed animals which did restore the rate of oxygen consumption (Vedpathak and Wagh, 2009), Vedpathak *et al.* (2011a) and Vedpathak *et al.* (2011b) and Mangesh Jadhav *et al.* (2012).A decrease in rate of oxygen consumption following injection of ganglionic extract to the intact animals which reached the normal intact control, confirms that the regulating link is not through the nervous input but possibly by neurosecretory. This contention can further be supplemented by the fact that even in intact control animals, as injection of extract of cerebral ganglia significantly decreases the rate of oxygen consumption.

Hence, it is concluded that, cerebral ganglia must possesses oxygen consumption controlling factor and which is neurosecretory. This integrity of these ganglia is essential in normal functioning of physiological activities of the bivalve molluscs. Similarly equivalent commercial hormones (progesterone and estradiol) also essentially playing a principle role in normal functioning of respiratory activity.

In the earth worm, *perionyx excavates*, the rate of oxygen consumption has been suggested to be under the influence of neurosecretory release of one or more hormonal agents from central nervous system (Nagabhushanam and Hanumante, 1977). The brain and subpharyngeal ganglia of earthworm have shown to be the oxygen inhibiting and elevating hormones respectively. The concept of hormonal control of oxygen consumption has been evidenced in number of poikilothermic organisms (Kale and Rao, 1973). In crab *Uca pugilator* two independent active hormones, controls the rate of oxygen consumption (1) Eyestalk factor regulating oxygen consumption and (2) The removal of moult inhibiting hormone which enhancing oxygen consumption (Silverthorn, 1973). In penaid prawn, *parapenaeopsis hadwickii*, eyestalk possesses a hormone which decreasing the rate of oxygen consumption (Nagabhushanam and Kulkarni, 1979).




(a) Control (normal), (b) Injection of cerebral ganglionic extract of same species to intact individuals, (c) injection of equivalent hormone progesterone to normal control and (d) injection of Estradiol to normal control

•-p<0.05, ••-p<0.01, •••-p<0.001

**Figure 1: Changes in the rate of oxygen consumption of fresh water bivalve, *Indonaia caeruleus* (Prashad, 1918) with injection of cerebral ganglionic extract and equivalent commercial hormones during summer**

In gastropod, *Onchidium verruculatum* removal of whole central nervous system or pleuropedal ganglia significantly inhibit oxygen uptake (Hanumante et al., 1980). Replacement of pleurovisceral ganglia in pleurovisceraloectomised gastropod restores the rate of oxygen consumption to the normal level.

In the present study, on freshwater bivalve, *Indonaia caeruleus*, it is possible that injection of their cerebral ganglionic extracts to intact animals could have resulted in initiation of the release of large quantities of serotonin and catacholamines as stated by Lubet (1970) in *Mytilus edulis*. Another exiting possibly that these neuro hormones after their entry into "milieuinterieur" may be enhancing the role of non –specific stressors (Gold and Ganong, 1977) or neuroendocrine transducers (Wurtman, 1972), there by indicating the endogenous neurosecretory hormones involved in regulation of oxygen consumption. This idea gives strength to the fact that the biogenic amines act as neurotransmitter to elicit the release of neurohormones from hypothalamic nuclei of vertebrate (Maclead and Lehmeyer, 1977) and probably also from those of invertebrates e.g. crustaceans (Fingerman et al., 1974) and bivalve molluscs (Mane et al., 1990). These neurohumors are capable of inducing changes in the neurosecretory cells in the cerebral and visceral ganglia of the bivalve mollusc (Kapoor, 1986)

Since the endogenous presence of these neurohormones in ganglia of bivalve molluscs as well as equivalent commercial hormones like progesterone and estradiol have already been established, regulation of oxygen consumption may be tentatively suggested as one of the physiological roles for these neurohumors in the metabolic economy of the bivalves.

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