

Effect of water depth on the hatching of eggs of three disease transmitting freshwater snails

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ABSTRACT. The eggs of three disease transmitting freshwater snails, *Lymnaea (Radix) luteola*, *L. (R.) acuminata* and *Indoplanorbis exustus*, were kept at different depths of water to detect effects on the development of embryos and hatching. For all three species, hatching decreased with increasing water depth. The eggs of *L. (R.) acuminata* and *I. exustus* failed to hatch when kept at a depth of 2.1 m or more those of *L. (R.) luteola* failed to hatch when kept at a depth of 2.4 m. The incubation period gradually increased with water depth.

INTRODUCTION

The freshwater snails *Lymnaea (Radix) luteola* Lamarck, *L. (R.) acuminata* Lamarck and *Indoplanorbis exustus* (Deshayes) are known intermediate hosts for some worm parasites such as *Fasciola gigantica*, *F. hepatica*, *Schistosoma indicum*, *S. nasalis*, *S. spindalis*, *Clinostomum giganticum* and others, causing diseases in man and his domestic animals (RAUT 1986; SUBBA RAO 1989). A number of studies (DURANI & CHAUDHURY 1967; ISLAM 1977; CHITRAMVONG *et al.* 1981; RAUT & GHOSH 1985; PARASHAR & RAO 1986; PARASHAR *et al.* 1986; RAUT & SAHA 1989; RAUT & MISHRA 1991, 1993; SAHA & RAUT 1992; RAUT *et al.* 1992; MISHRA & RAUT 1993) were effected with the aim of controlling the snail-borne diseases through the management of snail population. To achieve success, we still need more information on the biology and ecology of the vector molluscs.

These snails are found in large numbers in ponds and other freshwater bodies, in which they deposit eggs on a variety of substrates (fixed or not), normally near to the surface of the water. It could thus be expected that the development of the eggs could be hampered, if forced to take place in deeper water, for instance due to the heavy monsoon rainfall.

This work reports an experimental study of the effect of water depth on the hatchability of eggs of *L. (R.) luteola*, *L. (R.) acuminata* and *I. exustus* in a pond in Calcutta, West Bengal, India.

MATERIALS AND METHODS

Sexually mature specimens of *L. (R.) luteola*, *L. (R.) acuminata* and *I. exustus* were collected from the pond located in the Ballygunge Science College campus, Calcutta University, Calcutta, in the first week of December 1993. The species were then raised

separately, following the methods suggested by RAUT (1991), in 45x15x30 cm aquaria, in their water of origin, with a few aquatic plants (*Vallisneria*, *Ipomea* and *Lemna*) to provide a support for the snails to rest and lay eggs on. The molluscs were regularly fed with lettuce leaves. The snails started laying eggs within a few days.

The eggs were then returned under controlled conditions, as described here under, to the pond of origin of the snails. The experiments were initiated when the required number of eggs was available. At least 50 eggs were observed for each case.

The experimental set-up consisted in groups of three open-mouthed glass tubes (7 cm long and 2 cm diameter) tightened, at an interval of 15 cm, to a weighted 3 m nylon rope, rope secured to and hanging along a bamboo pole planted into the deepest part of the pond. The uppermost group of glass tubes was kept at the water surface (depth zero); the lowermost group being then at a depth of 270 cm. Series of three tubes at 19 predetermined depths could thus be raised or lowered in the water at will. Three such devices were used simultaneously providing three independent experiments and a total of 57 glass tubes. The experimental site of the pond was clean and exposed to direct sunlight for a daily period of 6 hours.

Plant parts bearing a recorded number of egg capsules, laid on the same day, of the three species of snails under study, were transferred into the tubes, which were then closed at both ends with 0.01 mesh nylon net. The eggs of the three species were kept separate for every tested depth. The eggs were examined, the hatched eggs counted and removed from the tubes at every 24 hours interval for a period of 40 days. The average hatching percentages was computed for eggs of each species, at each pre-set depth, in three sets of experiments.

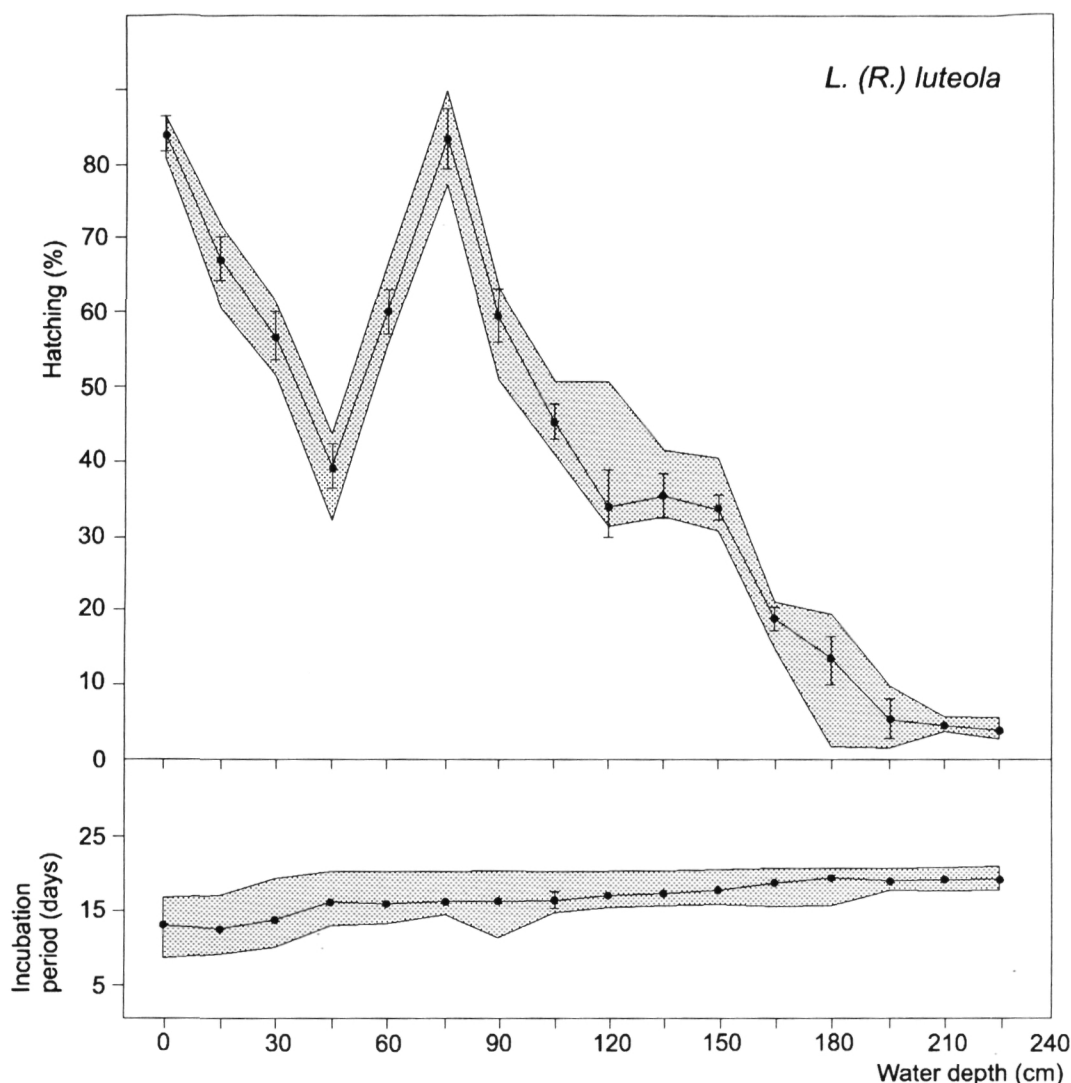


Figure 1. Hatching period and hatching percentages of the eggs of *L. (R.) luteola*. Range (shadowed area) and mean \pm S.E. (error bars not shown if very small).

RESULTS

At depth zero the incubation period of the eggs was 9-17 days for *L. (R.) luteola*, 11-19 days for *L. (R.) acuminata* and 9-14 days for *I. exustus*. The duration of the developmental period of the embryos and the percentages of eggs hatching varied both with the depth and with the species of the snails (Figures 1-3). Total failure to hatch was observed at 210 cm for *L. (R.) acuminata* and *I. exustus*, at 240 cm for *L. (R.) luteola*. The unhatched eggs of all the glass tubes were distorted and spoiled within 40 days.

Hatching time. The minimum hatching time is 9 days for the eggs of *L. (R.) luteola* and *I. exustus*, 10 days for *L. (R.) acuminata*. If kept in depths 0 to 15 cm [for *L. (R.) luteola*], 0 to 30 cm (for *I. exustus*) and 0 to 15 cm [for *L. (R.) acuminata*] only some of the eggs hatch in this minimum time while others require 10 to 14 days (for *I. exustus*), 10 to 17 days [for *L. (R.) luteola*]

and 11 to 17 days [for *L. (R.) acuminata*]. (Figures 1-3).

The maximum hatching time at depth zero was 14, 17 and 19 days for *I. exustus*, *L. (R.) luteola* and *L. (R.) acuminata*, respectively. For *L. (R.) luteola*, the maximum hatching time increased by one or two days at the depths of 15 to 30 cm, but never exceeded 20 days at any deeper location. At depths of 30 to 195 cm, the maximum hatching time ranged from 16 to 24 days and 20 to 25 days for *I. exustus* and *L. (R.) acuminata*, respectively.

Hatching of the eggs of the three snail species was thus considerably delayed by increasing the water depth. The minimum hatching time increased by 1 to 9 days for *L. (R.) luteola*, 1 to 7 days for *L. (R.) acuminata* and 1 to 5 days for *I. exustus*. The maximum hatching time increased by 2 to 10 days for *I. exustus*, 2 to 3 days for *L. (R.) acuminata* and 1 to 6 days for *L. (R.) luteola* (Figures 1-3).

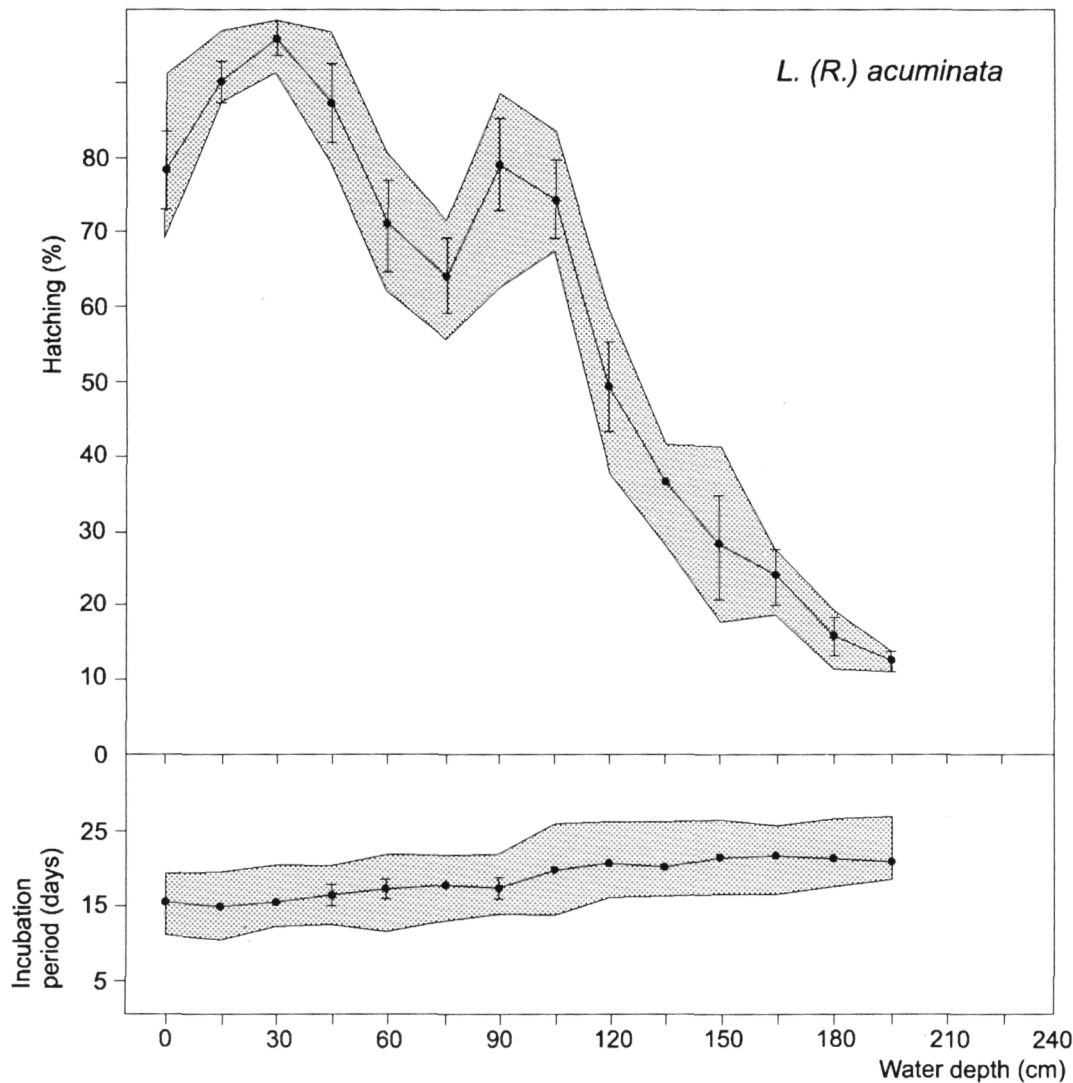


Figure 2. Hatching period and hatching percentages of the eggs of *L. (R.) acuminata*. Range (shadowed area) and mean \pm S.E. (error bars not shown if very small).

Hatching success. The percentage of eggs hatching varied with the depth of water. 84% of the eggs of *L. (R.) luteola* hatched when kept at depth zero, but only an average 2.6% when kept at 2.30 m. The same general trend is observed for the eggs of *L. (R.) acuminata* and *I. exustus*, for which the percentage of hatching falls to around 10% at the deepest depth tolerated (1.95 m).

DISCUSSION

It is to be noted that hatching time is a monotonous function of water depth, but hatching success is not. While the general trend of decreasing hatchability with increases depth is common to the three species, a small abnormality occurs around depths of 90 cm for *L. (R.) acuminata*. This corresponds to a larger peak of recovery around at around -90 cm for *L. (R.) luteola*

and a large, significant zone of relative success in the depth zone from -90 to -150 cm for *I. exustus*. The depth of the water plays a significant role in regulating the developmental process of the embryos but one should also note that the observed effect might be due to a number of factors such as pressure, intensity of light, temperature, pH, the amount of dissolved O_2 and the ionic concentrations of water at different depths.

In eggs laid in fresh water, a great discrepancy exists between the inner and outer osmotic pressure (RAVEN 1972). The envelopes and capsular membranes of the eggs of *Lymnaea* and *Biomphalaria* are permeable to water and inorganic ions (BEADLE 1969; RAVEN 1970). As a rule, the eggs are in osmotic equilibrium with water; they swell upon dilution of the medium and shrink when it is made hypertonic.

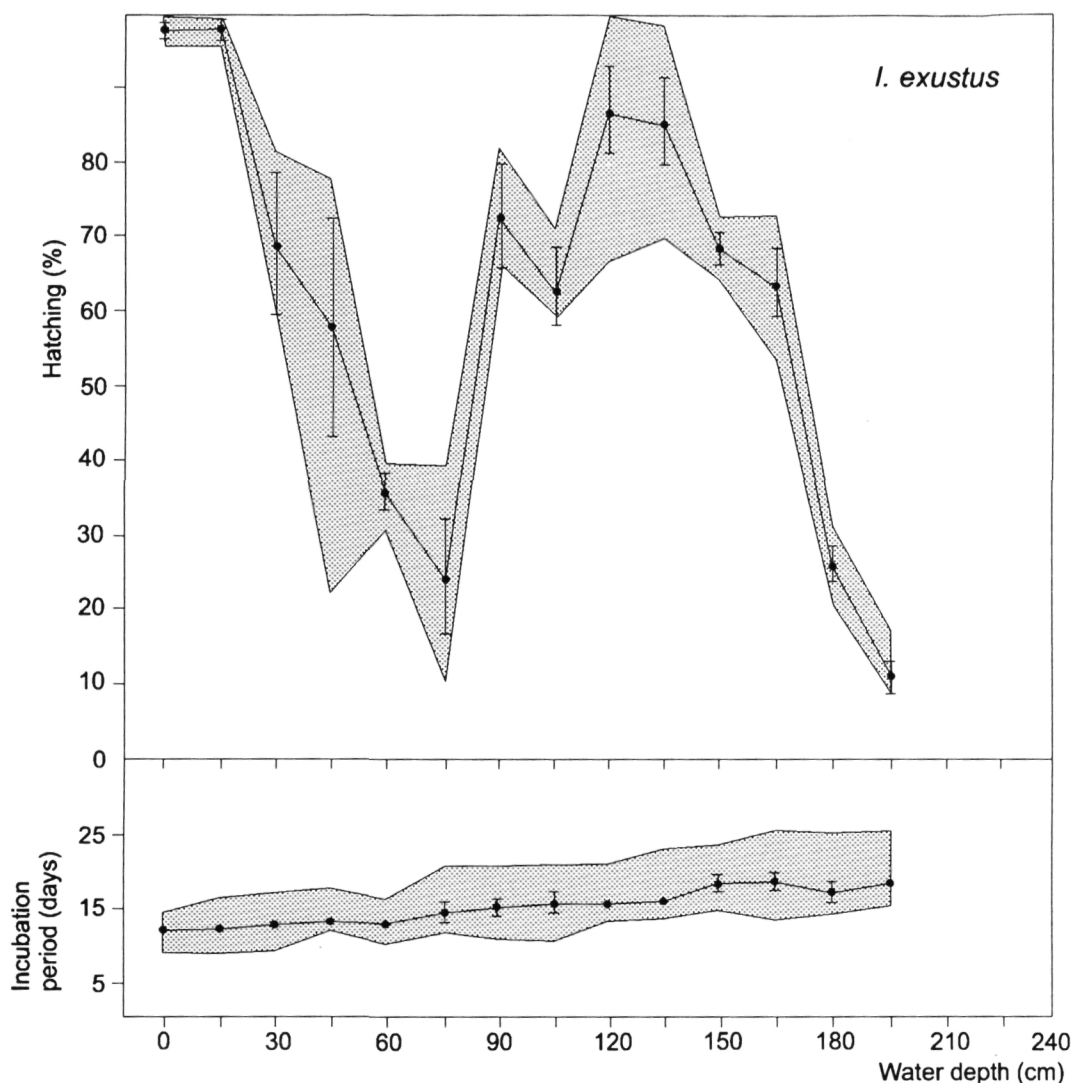


Figure 3. Hatching period and hatching percentages of the eggs of *I. exustus*. Range (shadowed area) and mean \pm S.E. (error bars not shown if very small).

We may expect to find in such eggs special mechanisms preventing excessive swelling (RAVEN 1972). This swelling is an osmotic phenomenon and its rate is regulated by the osmotic pressure of the medium (RAVEN & KLOMP 1946). The swelling of the eggs of *Lymnaea* continues after the onset of cleavage, even at a greatly increased rate (RAVEN *et al.* 1952; RAVEN 1966). This cannot be explained by the increase in surface area or a rise in osmotic pressure of the cells, but must be due to an increase in water permeability of the egg surface (RAVEN 1972). Incorporation of P^{32} into RNA shows a sharp increase during the third day of development of *Ilyanassa* eggs (RAVEN 1972). According to BRAHMACHARY *et al.* (1968), phosphorus-32 incorporation in *Lymnaea* begins in the uncleaved eggs, rises to a peak at the late trochophore stage, then declines throughout the veliger stage.

ROSS & HARRISON (1977) cultured eggs of the basommatophoran snails *Physa marmorata* and *Biomphalaria glabrata* in low concentrations of calcium, to determine effects on growth and development. In both species there was some development in media with 0.112 mg/l Ca^{++} but embryos did not hatch. 61% embryos of *P. marmorata* hatched in 0.22 mg/l Ca^{++} but those of *B. glabrata* required a level of 0.42 mg/l Ca^{++} to attain even a 31% hatch. Very low calcium levels induced marked effects on growth rate, embryo size and on hatching time taken in both species. These data indicate that a number of phenomena in the development of eggs imply ions exchange. Any obstacle to ions exchange is a danger for the eggs and invites different types of problems due to accumulation or release of different ions, and could inhibit the process of development, as it has been noted in case of the incorporation of P^{32} (BRAHMACHARY *et al.* 1968).

We have no data on ions concentrations neither in the water at the different depths that were tested, nor within the eggs; but we know that these ions concentrations are very much influenced by temperature and light. It is also known that temperature has a direct, significant impact on the development of eggs (DREGOLSKAYA 1986; SAHA & RAUT 1992). Pond water is clinograde, the amount of dissolved O₂ being maximum at the surface and gradually decreasing with increasing depth (GOLDMAN & HORNE 1983). It is thus likely that the eggs kept in deep waters suffer a deficiency of oxygen, inhibiting the development of the embryo.

Several factors being probably involved, understanding the mechanisms of the observed effects

still requires further study. As *L. (R.) luteola*, *L. (R.) acuminata* and *I. exustus* are very much involved with the spread of worm diseases any process reducing the density of these snail populations should be considered. Impairing the development of the eggs by controlling water depth could be an effective method.

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REFERENCES

- BEADLE, L.C. 1969. Salt and water regulation in the embryos of freshwater pulmonate molluscs 1. The embryonic environment of *Biomphalaria sudanica* and *Lymnaea stagnalis*. *J. exp. Biol.* 50: 481-489.
- BRAHMACHARY, R.L., K.P. BANERJEE & T.K. BASU. 1968. Investigations on transcription in *Lymnaea* embryos. *Exp. Cell. Res.* 51: 177-184.
- CHITRAMVONG, Y.P., E.S. UPATHAM & N. SUKHAPANTH. 1981. Effect of some physico-chemical factors on the survival of *Bithynia siamensis*, *Radix rubiginosa* and *Indoplanorbis exustus*. *Malacol. Rev.* 14: 43-48.
- DREGOLSKAYA, I.N. 1986. The asynchronism of hatching and the variability of heat resistance in juvenile pond snail (*Lymnaea fragilis*). *Zh. Obshch. Biol.* 47: 417-421.
- DURANI, M.Z. & A.H. CHAUDHURY. 1967. Studies on the life-cycle of *Indoplanorbis exustus* (Planorbidae: Gastropoda) under controlled conditions. *Pak. J. Vet. Sci.* 1: 14-16.
- GOLDMAN, C.R. & A.J. HORNE. 1983. *Limnology*. International Student Edition. McGraw-Hill International Book Company. Tokyo, Japan. xiii + 464 pp.
- ISLAM, K.S. 1967. A note on the life-cycle of *Indoplanorbis exustus* (Deshayes) under laboratory conditions. *Ind. J. Anim. Sci.* 47: 374-376.
- MISRA, T.K. & S.K. RAUT. 1993. Dynamic of population - dynamics in a medically important snail species *Lymnaea (Radix) luteola* (Lamarck). *Mem. Inst. Oswaldo Cruz*, Rio de Janeiro 88: 469-485.
- PARASHAR, B.D. & K. M. RAO. 1986. Effects of long-term exposure to low concentrations of molluscicides on a freshwater snail, *Indoplanorbis exustus*, vector of schistosomiasis. *Malacologia* 27: 265-269.
- PARASHAR, B.D., A. KUMAR & K.M. RAO. 1986. Role of food in mass cultivation of the freshwater snail, *Indoplanorbis exustus*, vector of animal schistosomiasis. *J. Moll. Stud.* 52: 120-124.
- RAUT, S.K. 1986. Snails and slugs in relation to human diseases. *Env. & Ecol.* 4: 130-137.
- RAUT, S.K. 1991. Laboratory rearing of medically and economically important molluscs. pp. 79-83. In: Director, Zoological Survey of India (ed.). *Snails, flukes and man*. Zoological Survey of India, Calcutta.
- RAUT, S.K. & UMA GHOSH. 1985. Egg capsules of some freshwater disease transmitting snails. *Env. & Ecol.* 3: 214-217.
- RAUT, S.K. & T.C. SAHA. 1969. The role of the water-bug *Sphaerodema annulatum* in the control of disease transmitting snails. *J. Med. Appl. Malacol.* 1: 97-106.
- RAUT, S.K. & T.K. MISRA. 1991. Effect of salinity on the growth of three medically important snail species. *Bull. Malacol. R.O.C.* 16: 67-74.
- RAUT, S.K. & T.K. MISRA. 1993. Influence of salinity on the breeding of three medically important freshwater snail species. *Malacol. Abh. Mus. Tierkd. Dresden.* 16: 173-176.
- RAUT, S.K., M.S. RAHMAN & S.K. SAMANTA. 1992. Influence of temperature on survival, growth and fecundity of freshwater snail *Indoplanorbis exustus* (Deshayes). *Mem. Inst. Oswaldo Cruz*, Rio de Janeiro 87: 15-19.
- RAVEN, C.P. 1966. *Morphogenesis: The analysis of molluscan development*. 2nd ed. Pergamon Press, London. xiii + 365 pp.
- RAVEN, C.P. 1970. The cortical and subcortical cytoplasm of the *Lymnaea* egg. *Int. Rev. Cytol.* 28: 1-44.

- RAVEN, C.P. 1972. Chemical embryology of Mollusca. pp.155-180 in: M. FLORKIN and B.T. SCHEER, eds. *Chemical Zoology*, Vol. VII, Mollusca. Academic Press, New York.
- RAVEN, C.P. & H. KLOMP. 1946. The osmotic properties of the egg of *Limnaea stagnalis* L. *Proc. K.ned. Akad. Wet. Amsterdam* 49: 101-109.
- RAVEN, C.P., J.J. BEZEM & J. ISINGS. 1952. Changes in the size relation between macromeres and micromeres of *Lymnaea stagnalis* under the influence of lithium. *Proc. K. ned. Akad. Wet. Amsterdam* 55C: 248-258.
- ROSS, J.F. & A.D. HARRISON. 1977. Effects of environmental calcium deprivation on the egg masses of *Physa marmorata* Guilding (Gastropoda: Physidae) and *Biomphalaria glabrata* Say (Gastropoda: Planorbidae). *Hydrobiologia* 55: 45-48.
- SAHA, T.C. & S.K. RAUT, 1992. Influence of temperature on hatching of eggs of some freshwater disease transmitting snails. *J. Taiwan Mus.* 45: 75-81.
- SUBBA RAO, N.V. 1989. *Hand Book : Freshwater molluscs of India*. Zoological Survey of India, Calcutta, xxiii + 289 pp.