
RESEARCH ARTICLES

J. Sci. Soc. Thailand 12 (1986) 23-30

EFFECTS OF SOME PHYSICO-CHEMICAL FACTORS ON THE HATCHING OF EGG MASSES AND ON THE SURVIVAL OF JUVENILE AND ADULT SNAILS OF *BULINUS (PHYSOPSIS) ABYSSINICUS*

MOHAMED A. DAGAL, E. SUCHART UPATHAM, MALEEYA KRUATRACHUE and VITHOON VIYANANT

Center for Applied Malacology and Entomology, Department of Biology, Faculty of Science, Mahidol University, Bangkok, Thailand

(Received 28 November 1985)

Abstract

*Effects of some physico-chemical factors--salinity, water temperature and hydrogen ion concentration--on the hatching of eggs and on the survival of juvenile and adult snails of *Bulinus (Physopsis) abyssinicus* were studied. All factors were found to affect the hatchability of egg masses and the survival of juvenile and adult snails. As the concentrations of sodium chloride solutions increased, the rates of hatching of egg masses and the survival rates of juvenile and adult snails decreased. The optimum temperatures for the hatchability of egg masses and the survival of snails were between 25°-30° C, respectively. The best pH at which the egg masses could hatch and the snails could survive well was 7.5. Below pH 6 and above pH 8, no eggs hatched and none of the snails survived.*

Introduction

Schistosomiasis is of a considerable public health significance in the Democratic Republic of Somalia. *Bulinus (Physopsis) abyssinicus* is the snail intermediate host of the only human schistosome known in the country, *Schistosoma haematobium*.^{1,2}

Infection occurs mainly in the valleys of Shebelli and Juba Rivers, South Somalia. The snail is generally regarded as the most vulnerable link in schistosome life cycle and, consequently, has been the main focus of attack in an attempt to reduce the transmission of infection³. This is done mainly by means of mollusciciding, a method which is usually expensive, has only a temporary effect and is liable to have undesirable side effects.

Ecological methods of control, where possible, would be more practical, although the initial costs might be high. These might include physico-chemical factors that might help to understand better the ecology of snails in breeding sites of particular hazard to community.

The objectives of this work were to study the effects of some physico-chemical factors (water temperature, hydrogen ion concentration and salinity) on the hatchability of egg masses and on the survival of juvenile and adult snails of *Bul. (P.) abyssinicus*.

Materials and Methods

A culture of *Bulinus (Physopsis) abyssinicus* from the Democratic Republic of Somalia has been established in the laboratory of the Center for Applied Malacology and Entomology, Faculty of Science, Mahidol University since 1978. The snails were fed with lettuce leaves continuously and their egg masses were collected daily. The eggs used were 24 h old. The initial stage of development and the number of eggs per mass were recorded prior to each experiment. The number of eggs hatched from each egg mass and the number of dead embryos were recorded daily. The results of all experiments were a mean of two replicates.

Juvenile and adult snails were selected from stock aquaria and washed thoroughly with dechlorinated water. The size of shell diameter was 5-7 mm for juvenile snails and over 7 mm for adult snails. Snails were placed in experimental containers kept under each experiment for 24 h and allowed to recover for another 48 h. After the recovery period, the mortality of snails was determined by probing at snail tentacles and head-foot portion and recorded. Every experiment comprised 2 replicate trials in the gradient.

Salinity

A graded series of nine sodium chloride solutions were used to test the hatchability of egg masses: control, 100, 200, 400, 800, 1600, 2000, 2400, 2800 and 3200 mg/l. For juvenile and adult snails, another graded series was set up: control, 100, 200, 400, 800, 1600, 2000, 2400, 2800, 3200, 3600, 4000, 4400, 4800, 5200, 5600, 6000, 6400, 6800, 7200 and 7600 mg/l.

Altogether, 1898 eggs, 840 juvenile and 840 adult snails were used.

Water temperature

The egg masses, juvenile and adult snails were exposed to various temperatures ranging from 5° to 40° C with 5° C intervals for 24 h in the case of snails and until they hatched or died for egg masses. At water temperatures of 5°, 10°, 15° and 20° C, egg masses and snails were placed in low temperature incubators; at 25° C, they were placed in an air-conditioned room; and at 30°, 35° and 40° C, they were placed in waterbaths.

Altogether, 1641 eggs, 320 juvenile and 320 adult snails were used.

Hydrogen ion concentration

Egg masses and snails were exposed to phosphate buffers with pH values ranging from 5 to 9 at one pH value intervals. The control groups were exposed to dechlorinated water with pH value of 7.5 for 24 h. All pH values were measured before and after the exposure period.

Altogether, 801 eggs, 240 juvenile and 240 adult snails were used.

Results

Salinity

The effects of sodium chloride on hatchability of eggs and survival of juvenile and adult snails, *Bul. abyssinicus* are shown in Table 1. The hatching rate of egg masses was highest in the control group (97.1%) and was inversely proportional to the increase of salinity. The maximum tolerated concentration for hatchability of eggs was 2800 mg/l (15.5%) and their lethal concentration was 3200 mg/l.

Salinity had no effect on survival of juvenile and adult snails in the range of sodium chloride concentration up to 5200 mg/l and 5600 mg/l, respectively. Between 4800 and 7600 mg/l for juvenile snails, and between 6000 and 7600 mg/l for adult snails, the mean percentage survival rates were inversely proportional to the concentration. Feeding habit, movement and other activities of juvenile and adult snails decreased with increased concentrations. The maximum tolerated concentration by which juvenile and adult snails could survive was 7200 mg/l (40%, 10%), and their lethal concentration was 7600 mg/l.

Water temperature

The effects of water temperatures on hatchability of eggs and survival of snails are shown in Table 2. At low temperatures (5°, 10° and 15° C), none of the eggs hatched. Similarly, the mean survival rate of juvenile and adult snails at 5° and 10° C was zero. As water temperatures increased up to 30° C, the mean percentage of hatching rate and survival of snails increased. The best hatching temperatures for eggs of *Bul. abyssinicus* were between 25° and 30° C (94.4%, 96.5%). At 35° C, none of the eggs hatched. The best survival temperatures for both juvenile and adult snails fell between 20° and 35° C (100–95%). At 40° C, none of the juvenile and adult snails survived.

Hydrogen ion concentration

The effects of pH values are shown in Table 3. At pH 5, the mean percentage of hatching rate of eggs was zero and all juvenile and adult snails were dead instantly when exposed. As pH values increased, the hatching rate and survival rate of snails increased. The best pH values for hatching of eggs and survival of snails fell between 7.5 and 8. Below pH 6 and above pH 8, no eggs hatched and none of the snails survived.

TABLE 1. EFFECTS OF SALINITY ON HATCHING OF EGGS AND SURVIVAL OF JUVENILE AND ADULT SNAILS.

Concentrations (mg/l)	No. of eggs hatched	(%)	No. of survival juveniles	(%)	No. of survival adults
	No. of eggs tested		No. of juveniles tested		No. of adults tested
Control	203/209 (97.1)		40/40 (100)		40/40 (100)
100	177/187 (94.6)		39/40 (97.5)		37/40 (92.5)
200	195/210 (92.8)		39/40 (97.5)		40/40 (100)
400	167/182 (91.8)		40/40 (100)		36/40 (90.0)
800	217/237 (91.5)		40/40 (100)		35/40 (87.5)
1600	188/215 (87.4)		40/40 (100)		38/40 (95.0)
2000	101/144 (70.1)		40/40 (100)		39/40 (97.5)
2400	58/110 (52.7)		40/40 (100)		35/40 (87.5)
2800	23/148 (15.5)		40/40 (100)		37/40 (92.5)
3200	0/256 (0)		40/40 (100)		40/40 (100)
3600	—		40/40 (100)		40/40 (100)
4000	—		40/40 (100)		39/40 (97.5)
4400	—		38/40 (95.0)		39/40 (97.5)
4800	—		40/40 (100)		39/40 (97.5)
5200	—		39/40 (97.5)		39/40 (97.5)
5600	—		37/40 (92.5)		40/40 (100)
6000	—		36/40 (90.0)		34/40 (85.0)
6400	—		33/40 (82.5)		33/40 (82.5)
6800	—		29/40 (72.5)		27/40 (67.5)
7200	—		16/40 (40.0)		4/40 (10.0)
7600	—		0/40 (0)		0/40 (0)

TABLE 2. EFFECTS OF WATER TEMPERATURES ON HATCHING OF EGGS AND SURVIVAL OF JUVENILE AND ADULT SNAILS.

Water temperatures (°C)	No. of eggs hatched		No. of survival juveniles		No. of survival adults	
	No. of eggs tested	(%)	No. of juveniles tested	(%)	No. of adults tested	(%)
5	0/268	(0)	0/40	(0)	0/40	(0)
10	0/246	(0)	0/40	(0)	0/40	(0)
15	0/213	(0)	21/40	(52.5)	21/40	(52.5)
20	94/284	(33.0)	40/40	(100)	40/40	(100)
25	106/111	(95.4)	40/40	(100)	40/40	(100)
30	279/289	(96.5)	39/40	(97.5)	39/40	(97.5)
35	0/230	(0)	39/40	(97.5)	38/40	(95.0)
40	—		0/40	(0)	0/40	(0)

TABLE 3. EFFECTS OF pH VALUES ON HATCHING OF EGGS AND SURVIVAL OF JUVENILE AND ADULT SNAILS.

pH values	No. of eggs hatched		No. of survival juveniles		No. of survival adults	
	No. of eggs tested	(%)	No. of juveniles tested	(%)	No. of adults tested	(%)
5	0/100	(0)	0/40	(0)	0/40	(0)
6	132/147	(89.7)	37/40	(92.5)	36/40	(90.0)
7	126/136	(92.6)	38/40	(94.8)	38/40	(95.0)
7.5	116/121	(95.8)	40/40	(100)	40/40	(100)
8	138/145	(95.1)	40/40	(100)	36/40	(90.0)
9	0/152	(0)	0/40	(0)	0/40	(0)

Discussion

It is apparent from this study that salinity, water temperature and hydrogen ion concentration all have effects on the hatching of eggs and the survival of juvenile and adult snails of *Bul. abyssinicus*.

Salinity seems to have more effect on hatching of eggs than survival of snails. The hatching rate decreased as there was an increase in salinity concentration up to 3200 mg/l, at which none of the eggs hatched. This is in close agreement with that of Davis⁴ who studied the survival of clams. Hatching of egg masses appeared to be depressed by the increase of sodium chloride concentration. The egg masses exposed to sodium chloride solutions ranging from control to 1600 mg/l did not show any kind of abnormality in their capsules, in their process of hatching and development, but those exposed beyond the above range showed swelled capsules, especially the egg masses exposed in 2800 to 3200 mg/l. The layers of capsule materials were separated and appeared thick and dense. According to Allen⁵ who studied the effects of salinity in clams, it could be explained that as the clams progressed from dilute to a more saline environment, there was a loss of water. Similarly, the egg masses of *Bul. abyssinicus* might lose some of their water content as the concentration of salinity increased.

Both juvenile and adult snails of *Bul. abyssinicus* could tolerate salinity concentrations up to 6000 mg/l and their lethal concentration was 7600 mg/l. These results are contrary to the work of Descheins⁶ on *Bul. contortus*. According to Descheins⁶, the maximum tolerated salinity concentration of *Bul. contortus* was 2123 mg/l, and their lethal concentration was 3500 mg/l. It seems that *Bul. abyssinicus* could tolerate high salinity and this may be attributed to its ecology. *Bul. abyssinicus* are found mainly in the valleys of the Shebelli and Juba Rivers, South Somali. These two rivers originate from Ethiopian highlands where sodium chloride deposition is very high. Therefore, *Bul. abyssinicus* had probably been adapted to this nature of ecology as time had passed. Hence, both juvenile and adult snails could resist a very high range of salinity.

Water temperatures were found to affect the hatching of eggs and the survival of juvenile and adult snails of *Bul. abyssinicus*. At 5°C, there was no development of eggs, but at 10 and 15°C, there was an initial development although they did not hatch. This result is in consistent with that of Gordon *et al.*⁷ They found that, under laboratory condition, no spawning was observed at 15°C. However, the egg masses of *Bul. abyssinicus* started to hatch at 20°C. At 35°C, none of the eggs hatched. The high temperature had caused the high abnormalities in the embryos until they became inviable, so the eggs became degenerate and did not hatch. In addition, Malek⁸ pointed out that there was a relationship between temperature and oxygen consumption, the latter increased as the temperature rose. Hence, the egg masses of *Bul. abyssinicus* which failed to hatch at 35°C may be due to degeneration and reduction of dissolved oxygen in water due to higher temperature.

The optimal range of temperature for survival of juvenile and adult snails is between 20 to 35 °C. None of the snails survived at above or below this range. A rise in temperature causes metabolism to accelerate and accumulate harmful metabolites in ectothermic animals such as snails. In addition, it also promotes membrane permeability which may disturb the distribution of ions and small molecules on either side of the membrane. Therefore, the rise in temperature to 40 °C may have resulted in the death of all juvenile and adult snails. In addition, a fall in temperature from 15 to 5 °C apparently disturbed certain aspects of the snail physiological function, and this may have been associated with slow dehydration of the cells and cause the entire physiological system to stop so that death occurs.

The hatching of eggs and survival of juvenile and adult snails, *Bul. abyssinicus* could occur at pH 6–8. In all cases, the optimal pH was 7.5. This result is in close agreement with the work of Harrison and Agnew¹⁰ who found that some temperate species of freshwater molluscs such as *Radix natalensis* (Krauss), *Pseudosuccinea columella* (Say) and *Bul. tropicus* (Krauss), appeared to prefer alkaline water (pH 8.5).

Although hydrogen ion concentration is rarely a factor limiting the distribution of snail⁸, it highly influences the hatching and survival of *Bul. abyssinicus*. Below pH 5 and above pH 8, the physiological functions of the animal are disturbed. However, the combined effects of other factors correlated with pH alkali reserve, CO₂ content, sunlight, photosynthesis with its active removal of CO₂, and production of O₂, and the character of the substratum are more important than the pH alone.^{8,11}

Acknowledgement

This investigation was supported by the United Nations Development Programme/World Bank/World Health Organization Special Programme for Research and Training in Tropical Diseases.

References

1. Koura, M., Upatham, E.S., Awad, A.H. and Ahmed, M.D. (1981). Prevalence of *Schistosoma haematobium* in Koryole and Merca Districts of the Somali Democratic Republic. *Ann. Trop. Med. Parasit.* **75**, 53–61.
2. Upatham, E.S., Koura, M., Ahmed, M.D. and Awad, A.H. (1981). Studies on the transmission of *Schistosoma haematobium* and bionomics of *Bulinus (Physopsis) abyssinicus* in the Somali Democratic Republic. *Ann. Trop. Med. Parasit.* **75**, 63–69.
3. World Health Organization. (1981). Molluscicides. *Wld. Hlth. Org. Tech. Rep. Ser.* **214**, 1–50.
4. Davis, H.C. (1958). Survival and growth of clam and oyster larvae at different salinities. *Biol. bull.* **114**, 296–307.
5. Allen, K. (1961). The effects of salinity on the amino acids concentration in *Rangia cuneata* (Pelecypoda). *Biol. bull.* **121**, 420–424.
6. Descheins, R. (1954). Incidence de la mineralisation de l'eau sur les mollusques vecteurs des bilharzioses. Consequences pratiques. *Bull. Soc. Path. Exot.* **47**, 915–929.

7. Gordon, R.M., Davey, T.H., and Peaston, H. (1934). The transmission of human bilharziasis in Sierra Leone, with an account of the life-cycle of the schistosomes concerned, *S. mansoni* and *S. haematobium*. *Ann. Trop. Med. Parasit.* **38**, 323-418.
8. Malek, A.E. (1958). Factors conditioning the habitat of bilharziasis intermediate hosts of the family Planorbidae. *Bull. Wld. Hlth. Org.* **18**, 785-818
9. Wieser, W. (1973). Temperature relations of ectotherms: A speculative review. In: Wieser, W. (Ed.), *Effects of temperature on ectothermic organisms*. Springer-Verlag, Berlin, Heidelberg, New York. 298 pp.
10. Harrison, A.D. and Agnew, J.D. (1962). The distribution of invertebrates endemic to acid streams in Western and Southern Cape province. *Ann. Cape Prov. Mus. Nat. Hist.* **121**, 273-291.
11. Welch, P.S. (1952). *Limnology*, McGraw-Hill Book Company, New York, U.S.A. 538 pp.

บทคัดย่อ

จากการศึกษาเกี่ยวกับผลกระทบจากปัจจัยสภาวะแวดล้อมที่มีต่อการฟักเป็นตัวของไข่หอยและการออกรูดของหอยตัวอ่อนและตัวเต็มวัย *Bulinus (Physopsis) abyssinicus* ซึ่งเป็นหอยที่เป็นพาหะของพยาธิใบไม้ในเลือด *Schistosoma haematobium* ในทวีปแอฟริกา พบว่าปัจจัยสภาวะแวดล้อม ได้แก่ ความเป็นเกลือ อุณหภูมิของน้ำ และ pH ต่างก็มีผลต่อการฟักตัวของไข่หอยและการออกรูดของหอยชนิดนี้ทั้งสิ้น น้ำที่มี NaCl ปริมาณสูง 2,800 ppm, 5,200 ppm และ 5,600 ppm จะทำให้อัตราการฟักตัวของไข่ และอัตราการออกรูดของหอยตัวอ่อนและตัวเต็มวัยลดลงตามลำดับ อุณหภูมิที่เหมาะสมต่อการฟักตัวของไข่หอยและการออกรูดของหอยคือ 25 - 30°C ค่า pH ที่ดีที่สุดสำหรับการฟักตัวของไข่และการออกรูดของหอยชนิดนี้คือ 7.5 ถ้าค่า pH สูงหรือต่ำกว่านี้ไข่จะไม่ฟักเป็นตัวและหอยจะตายเป็นส่วนใหญ่