Rate of Filtration by *Perna viridis* Pre-exposed to Heavy Metals*

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Perna viridis exposed to different concentrations of copper and zinc for varying periods and then the rate of filtration estimated under metal free culture conditions. The concentrations under which the animals were maintained before the experiments were 0.025 to 0.150 p.p.m. of zinc and 0.005 to 0.08 p.p.m. of copper.

The filtration rate of bivalves is known to be influenced by the presence of pollutants apart from salinity, temperature, dissolved oxygen and concentration of suspended matter (Cole & Hepper, 1954; Badman, 1975). Most of the heavy metals, whether essential or otherwise, are potentially toxic to living organisms and are known to function as enzyme inhibitors above their optimum levels (Vallee & Walker, 1970). Among such heavy metals studied, copper has received considerable attention. The inhibitory effects of copper, silver and zinc on the filtering ability of mussels have been reported (Abel, 1976; Mathew & Menon 1984; Murthy, 1982). Abel (1976) working on *Mytilus edulis* reported that copper at 0.15 mg/l concentration reduced the filteration rate by 50%. Copper *in vitro* directly inhibited the ciliary action of *Mytilus edulis* (Brown & Newell, 1972).

Materials and Methods

Perna viridis and seawater were collected from an unpolluted region near the Someswara rocky shore (12° 47′ N; 74° 51′E). Seawater collected in polythene carboys was kept for aging (3–4 days) and filtered before use. Prior to use, the mussels were kept under laboratory conditions for 24 h (temperature 30 ± 1°C, salinity 33 ± 1‰, oxygen 4.8 – 5.3 ml/l, pH 8.3 – 8.4). During the course of the experiment the test animals were not fed.

Zinc chloride ($ZnCl_2$ $2H_2O$) and copper sulphate ($CuSO_4$ $5H_2O$) were the source of zinc and copper respectively. The experimental vessels were rectangular glass jars of 500 ml capacity containing 400 ml of test solution prepared in aged seawater. The rate of filtration was assessed by employing the dye clearance technique. This technique involves additoin of a known concentration of neutral dye (Neutral red 0.01%) to the test solution and allowing the animals to clear the dye. The reduction of dye concentration at fixed intervals was estimated colorimetrically with the help of a Klett-Summerson photo electric colorimeter using a blue filter (No. 42). Filtration rate was estimated using Quyle's (1948) equation, and the results are expressed as quantity of seawater (in ml) filtered by an individual in one hour.

P. viridis were exposed to various concentrations of zinc (0.025– – 0.150 p.p.m.) and copper (0.005 – 0.080 p.p.m) individually for a period varying from 24 h to 96 h. They then were transferred to clean seawater every 24 h and the rate of filtration was observed for a period of 6– 10 h. The frequency of reading was every 2 h. The basic idea was to assess whether exposure for a shorter duration would effect the rate of filtration even if the animals were transferred to clean seawater.

Results

In the case of animals pre-exposed for 24 h in various concentrations of zinc

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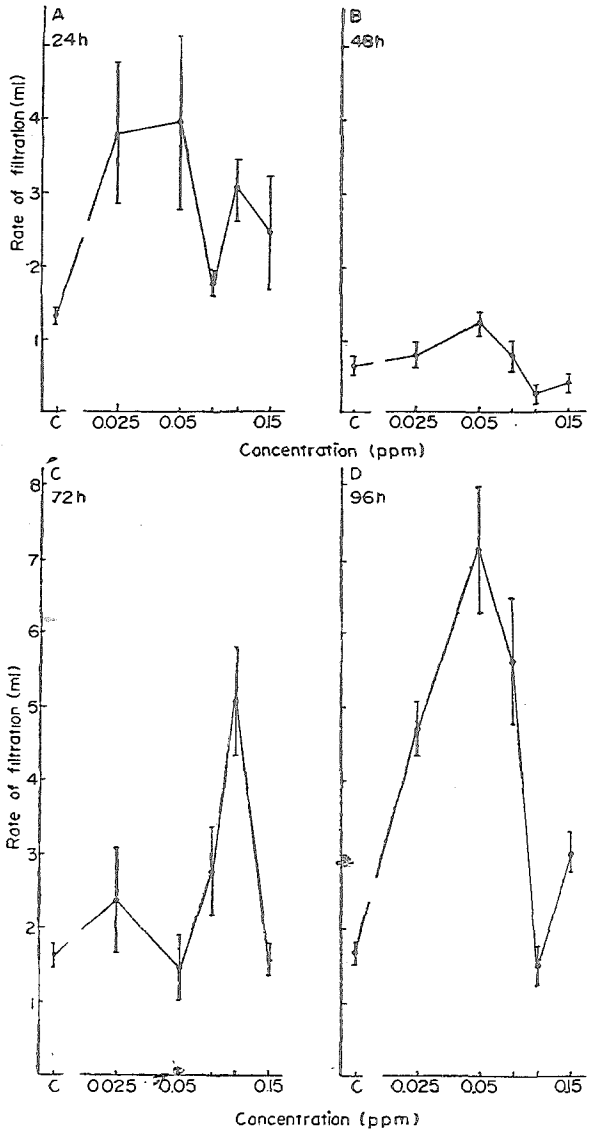

Fig. 1. A–D. Average rate of filtration (ml/h/individual) by *Perna viridis* pre-exposed to 24 h to 96 h to various concentrations of zinc. Vertical bars show standard deviation.

(0.025 to 0.150 p.p.m.), there was no clear cut variation in filtration rate. However, all the animals although in clean seawater showed an increase in filtration rate compared to those of control (Fig. 1a). Reduction in filtration rate was observed in animals pre-exposed for 48 h under varying zinc concentrations (Fig. 1b). During the course of the experiment animals pre-exposed to a few concentrations did not even filter for a period of 2 h while those pre-exposed to 0.10 and 0.150 p.p.m. of zinc filtered less quantity of water. Those test individuals when pre-exposed for 72 h and 96 h under various concentrations filtered more quantity than those in control. Here also between concentrations, there was no clear cut evidence to assume concentration controlled variation in filtration (Fig. 1c and 1d).

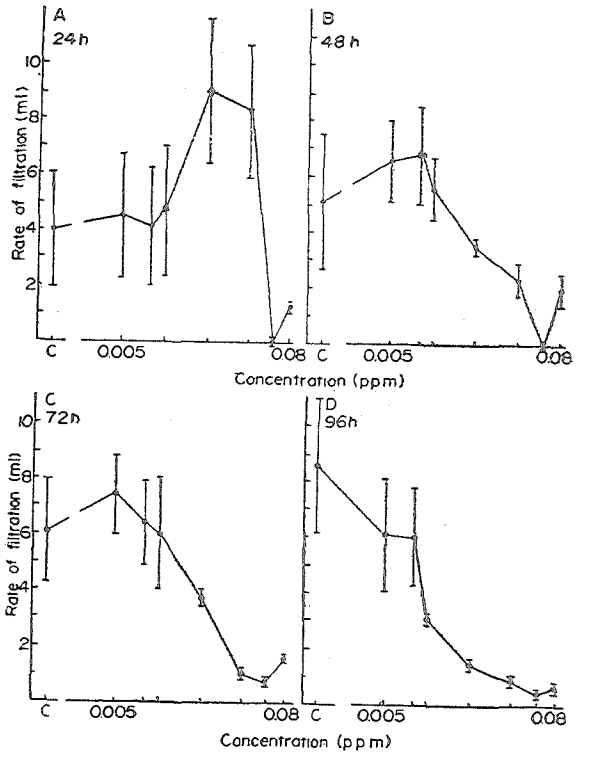

Fig. 2. A–D. Average rate of filtration (ml/h/individual) by *Perna viridis* pre-exposed to 24 h to 96h to various concentrations of copper. Vertical bars show half standard deviation.

Among those animals which were pre-exposed for a period of 24 h to copper (0.005 to 0.08 p.p.m.), only those which experienced 0.005 to 0.04 p.p.m. of copper stress there was an increase in filtration rate, whereas those pre-exposed to 0.06 and 0.08 p.p.m. of copper filtered less quantities of water (Fig. 2a). Although this trend was maintained in animals pre-exposed for 48 h (Fig. 2b) and 72 h (Fig. 2c) of copper stress, those who experienced 96 h duration of copper stress filtered decidedly less quantity of water (Fig. 2d).

Discussion

Rate of filtration and oxygen consumption are directly correlated to the valve closure mechanism of bivalves. Working

on *Mytilus* and *Crassostrea,* Shumway (1977) reported that in fluctuating salinity these two genera may exhibit full valve closure for a considerable amount of time and this could be as long as 96 h in *Mytilus edulis.* In the case of *Mytilus edulis,* Abel (1976) reported that the dye extraction efficiency may continue to change during test as the dye concentration decreases. Since filtration rates are assessed by dye clearance than by the actual water flow methods, it is likely that method of testing influences the results. Although shell movement could affect propulsion, the presence of excessive mucus on the gill might reduce the actual amount of dye solution contacting the gills. This might mean that the water entering the valves goes out without the dye being filtered giving a higher dye concentration irrespective of water propulsion. In the present experiments all the animals were pre-exposed to varous concentrations of zinc and copper and then tested for filtration efficiency in clean seawater. Therefore the results obtained are indicative of effects manifested by pre-exposure to the toxicants. Phillips (1977) suggested that the possibility of the pollutant itself eliciting valve closure (decreasing filtration rate) is more likely. The present results clearly show that a pre-exposure for 96 h at concentration as high as 0.15 p.p.m. of zinc might result in decrease in the rate of filtration, even if the animals are removed to fresh seawater. This probably shows that apart from inducing valve closure upon direct exposure, the quantity of zinc that would have entered the muscle tissue also induce reduction in filtration rate in the case of *Perna viridis*

The previous investigations have shown that copper generally influences reduction in the rate of filtration in *Perna viridis* and *Modiolus* sp., In the case of *Perna viridis,* filtration rate was reduced to 50% when exposed to 0.043 p.p.m. copper (Murthy, 1982). During the present investigation also filtration rate was reduced to 25–15% efficiency in concentrations ranging from 0.04 to 0.08 p.p.m. copper, although these animals were tested in clean seawater after exposure to various copper concentrations for durations ranging from 24 to 96 h. The present results clearly show that the pre-exposure did influence the rate of filtration, although the animals were maintained in unpolluted waters. The exact mechanisms of metal affecting filtration capacity is not known. The suppression of ciliary activity by copper ions may result in reduction in filtration rate. The present findings show that temporary exposure, to fresh seawater will not result in regaining normal filtration rate by the copper pre-exposed *Perna viridis* and also clearly indicate that certain damage is done to the filtering mechanism and valve closure mechanism of this species which could be of irrevocable nature as the concentrations employed were in the near abouts of LC50 values of the concerned metals.

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References

Abel, P.D. (1976) *Mar. Pollut. Bull.* **7,** 228

Badman, D.G. (1975) *Comp. Biochem. Physiol.* **51**A, 741

Brown, B. & Newell, R.C. (1972) *Mar. Biol.* **16,** 108

Cole, H.A. & Hepper, B.T. (1954) *J. Cons.* **20,** 197

Mathew, R. & Menon, N.R. (1984) *Mahasagar.* **17,** 183

Murthy, C.K. (1982) *Combined Toxicity of Silver and Copper on a Few Marine Invertebrates and Ecological Monitoring of the Inshore Waters of the Arabian Sea Off Mangalore.* pp. 274, M.F.Sc. Thesis, University of Agricultural Sciences, Bangalore

Phillips, D.J.H. (1977) *Mar. Biol.* 43, 283

Quyle, D.B. (1948) *Biology of Venurupis pullastra* (Montogu) Dissertation, University of Glasgow, pp. 62 (mimeograph).

Shumway, S.E. (1977) *Mar. Biol.* 4, 153

Vallee, B.L. & Walker, W.E.L. (1970) In: *The proteins,* (Neurath, H. ed.) Vol. 5, *Metalloproteins,* Academic Press, New York and London