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INFLUENCE OF pH ON UPTAKE AND BIOACCUMULATION OF RADIOCESIUM (¹³⁷Cs) IN THE GREEN MUSSELS *PERNA VIRIDIS*

*A. MASHIATULLAH, M.S. KHAN, T. JAVED, R.M. QURESHI¹, F. CHAUGHTAI² and M.Z. CHAUDHARY

Isotope Applications Division, Directorate of Technology, PINSTECH, P.O. Nilore, Islamabad, Pakistan

¹Directorate of Coordination, PINSTECH, P.O. Nilore, Islamabad, Pakistan

²Centre of Excellence in Marine Biology, University of Karachi, Karachi, Pakistan

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Radiotracer experiment was conducted to assess the influence of pH on uptake rate and bioaccumulation of ¹³⁷Cs from dissolved phase in soft parts of green mussels (*Perna viridis*) collected from Karachi Coast, under controlled laboratory condition. Green mussels of size 5–7 cm were subjected to radiocesium activity of 24 KBq L¹ under three pH levels i.e., 7.5, 8 and 8.5. The uptake of ¹³⁷Cs was monitored for a period of seven days. The results showed that uptake and bioaccumulation of ¹³⁷Cs in mussels was dependent on pH levels. Higher bioaccumulation factors and uptake rates were found at pH 8.

Keywords: Mussels, pH, Cesium, Bio-accumulation, Uptake, Karachi

1. Introduction

The green-lipped mussel *Perna viridis* is widely distributed in tropical and subtropical Asia [1, 2]. These are extensively used in monitoring programs in the marine environment due to their ability to concentrate pollutants to several orders of magnitude above ambient levels in sea water [3]. Mussels possess many characteristics that make them an ideal pollution indicator [4]. Specifically, these filter feeders tend to accumulate pollutants, especially metals, in their tissues and their body burdens of trace metals have often been used to reflect levels of contamination in surrounding waters. Mussels are long lived and easily sampled and their sedentary nature means that their geographical relationship to a pollution source can be easily ascertained. Mussels, therefore, exhibit desirable characteristics to serve as sentinel organisms. Many studies have demonstrated that over extended periods of time, mussels sequester and may accumulate heavy metals in their shells [5] and soft tissues [6-9]. The importance of bivalve mollusks in pollution impact studies is shown by the magnitude and longevity of the International

¹³⁷Cs (half life of 30.2 years) is an important fission product occurring in fallout from nuclear power tests and in effluent from nuclear reactors, the accumulation of cesium by aquatic organisms has assumed considerable significance. Marine organisms generally concentrate cesium 3-30 times over the level in the surrounding waters but in related fresh water species, concentration factors may be higher by two or three orders of magnitude [13]. Cesium has been shown to accumulate in both terrestrial and aquatic food chains [14]. Concern for radiocesium also arises because of its potential transfer within aquatic food webs, particularly its potential biomagnification at level hiaher trophic [15]. Uptake and bioaccumulation of ¹³⁷Cs in bivalves depends on many environmental factors such as salinity, temperature and pH. ¹³⁷Cs uptake at different salinity and temperature regime by green mussels has been carried out in our laboratory [16, 17]. In the present investigation ¹³⁷Cs uptake was studied in P. viridis at different pH levels. The objective of the present study was to determine the influence of one of the two key parameters, namely pH

* Corresponding author : azhar@pinstech.org.pk

Mussel Watch Programme [10-12].

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regimes, on the capacity of the green mussel *Perna viridis* to concentrate ¹³⁷Cs known to be possible contaminants of concern in the Karachi area. The pH ranges tested were selected to represent the ranges actually encountered in the coastal waters of Karachi and the experimental work was carried out using state-of-the-art nuclear techniques and gamma-emitting radiotracers of the ¹³⁷Cs.

2. Material and Method

2.1. Sampling

Green mussels (Perna viridis) were collected from the same bed and at same tidal level from Karachi Coast in February 2009. All the collected mussels had a size between 5 and 7 cm. The collected organisms were placed in plastic bags containing seawater from the sampling location and brought back to the laboratory in PINSTECH, Islamabad for acclimation. Mussel shells were cleaned of their fouling organisms and were distributed in three aquariums. They were acclimated to laboratory conditions (closed circuit aguaria, temperature 29 ± 1°C, salinity 35 ppt) for one week prior to experiments and fed daily with a mixture of Ankistrodesmes spp. and Navicula spp. of phytoplankton.

2.2. Experimental setup

All experiments were carried out using 0.22 μ filtered seawater in 8-L polyvinyl aquaria equipped with a small size circulation pump and an air pump. pH of seawater was 8.0. pH was adjusted to 7.5 by adding hydrochloric acid (HCl) and 8.5 by adding sodium hydroxide (NaOH) pallets. Mussels were acclimated to the selected pH levels i.e. 7.5, 8 and 8.5 for a week prior to the actual experiment and 50 mussels were placed in each aquarium. The experimental seawater of each aquarium was spiked with μ L quantities of ¹³⁷Cs (as cesium chloride in 1M hydrochloric acid) to reach a final activity of 24 kBq L⁻¹ [16].

2.3. ¹³⁷Cs analysis

At different times, water samples and three mussels were collected from each aquarium. The organisms were briefly rinsed with clean seawater, blotted dry, weighed, and dissected into shells and soft parts. The dissected parts were weighed (wet wt.) and radioanalyzed using a Triathler Multilabel Tester (Nal detector having relative efficiency >45 % with ¹²⁵I) for 5 minutes. The activities of all the samples were compared with standards of appropriate geometry and corrected for physical decay. The counting times were adjusted to yield a propagated counting error of <5% [16, 17].

2.4. Uptake rate and ¹³⁷Cs bioconcentration factor calculation

Bioconcentration Factor (BCF) is taken as the ratio of concentration of radionuclides in flesh wet weight (Bq gm⁻¹) and the seawater (Bq mL⁻¹) and expressed as L Kg⁻¹. By applying the BCF concept to marine organisms, it is assumed that the metal concentration in the organisms is at steady state with the concentration in the surrounding water and that the uptake of the metal is in proportion to its concentration in the seawater [18]. The uptake rate was calculated by slope of the linear regression between ¹³⁷Cs concentration in mussels and time of exposure and it is expressed as change in BCF over time (μ g g⁻¹h⁻¹).

3. Results and Discussion

The uptake kinetics of ¹³⁷Cs was determined in the soft tissues of mussels exposed for 168 hrs under different pH conditions (7.5, 8 and 8.5). Significant pH dependence of 137 Cs uptake is depicted in Fig. 1. The uptake (slope of regression bioaccumulation and between time) was considerably affected by pH levels. Maximal values of the ¹³⁷Cs uptake were observed at 8.0 with significant decreases at 7.5 and 8.5. The ratio of ¹³⁷Cs uptake rate ($\mu g g^{-1} h^{-1}$) in flesh of Perna viridis was found to be 1.7:4.3:1.0 at 7.5, 8 and 8.5 respectively after end of the experiment.

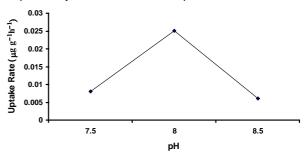


Figure 1. Uptake rate of Cs-137 in Perna viridis at different pH levels

¹³⁷Cs bioconcentration factor (BCF), also known as bioaccumulation factor (BF), was calculated as the ratio of ¹³⁷Cs concentration in the mussels to ¹³⁷Cs concentration in the dissolved phase. ¹³⁷Cs BCF in the mussel flesh at pH 7.5, 8 and 8.5 through dissolved phase is shown in Fig. 2. BCF was low at pH 8.5 as compared to 7.5 and 8. Uptake of ¹³⁷Cs from dissolved phase by P. viridis *shown* in Fig. 2 is time dependent process. It appears to be a two phase process at pH 7.5 and 8.5, characterized by the first rapid phase lasting 32 hrs followed by the second slow phase, reaching equilibrium within several hours. The second phase of ¹³⁷Cs uptake is quite slow after 32 hrs and almost equilibrium is attained. However at pH 8.5 ¹³⁷Cs uptake is appear to be a three phase process, an initial rapid uptake upto 32 hrs, 2nd phase is characterized by a steady state condition from 32 to 48 hrs and in third phase an increase in uptake at the end of experiment.

Table 1. Uptake rate and initial sorption rate of ¹³⁷Cs in *Perna viridis* flesh at different pH levels.

| pH Levels | Uptake rate (µg g ⁻¹ h ⁻¹) | Initial sorption (µg g⁻¹) |
|--------------|--|------------------------------|
| 7.5 | 0.012 | 1.17 |
| 8 | 0.030 | 0.746 |
| 8.5 | 0.007 | 0.804 |

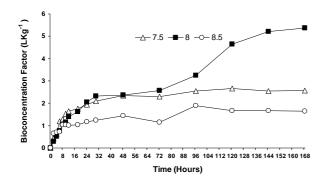


Figure 2. Bio concentration factor of ¹³⁷Cs by *Perna viridis* at different pH levels.

It is well known that organisms living in lower aquatic organism will show a positive response in environment close to their natural environment. The pH values of Karachi seawater are almost 8.0 that's the best possible explanation of higher BCF at pH 8.0 in the present study.

Various mechanisms can be proposed to explain pH effects on uptake of metals. Change in metal speciation and physiological conditions of the mussels such as the pumping rate, or change in cell volume and permeability known to be effected by pH change [19-21]. pH also influences directly the concentration of free metal ion, which is the most bio-available chemical species [22].

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However, changes in concentration of the major cations in seawater can also influence metal uptake by changing the permeability of the epithelial structures, competing for 140 binding sites with the apical membrane surfaces, and decreasing metal transfer from the epithelium to the blood with increasing intracellular levels of calcium [23-26]. Therefore, the influence of pH on the ¹³⁷Cs influx rate is probably due to the combined effects of speciation change (related to change in chloride anions) and change in binding competition (related to major cations). The uptake rate generally decreases until a steady state is reached between the metal in the water and in organism tissues. In larger organisms, internal tissues are often isolated from the surrounding water, with longer equilibration times for surface metal sorption from water (days to weeks) compared to small species such as mussels. The importance of the initial component of uptake depends to some extent on the surface characteristics of the organism. Hard-shelled, calcareous animals can deposit appreciable amounts of metal in the shell during growth, whereas soft-body organisms with no hard, external covering are able to equilibrate their internal tissues more rapidly.

4. Conclusions

The present study has clearly shown that bioconcentration of dissolved ¹³⁷Cs in the soft tissues of the mussels (*Perna viridis*) is significantly affected by changes in pH. Therefore, this parameter can be used to understand and predict metal bioaccumulation in natural conditions and for interpreting reliably the data of field monitoring studies. Furthermore, the ability of mussels to accumulate radiocesium in soft body parts can also be utilized for bio monitoring studies along Karachi Coast.

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