PUTATIVE BIOINDICATOR OF $^{137}$Cs IN PERN A VIRIDIS

Alejandro Q. Nato, Jr., Eliza B. Enriquez', Ariel T. Ortiz', and Custer C. Deocaris

1 Health Physics Research Section, Philippine Nuclear Research Institute
   Commonwealth Avenue, Diliman, Quezon City
2 The Marine Science Institute, University of the Philippines, Diliman, Quezon City
3 Cancer Research and Radiation Biology Laboratory, Philippine Nuclear Research Institute
   Commonwealth Avenue, Diliman, Quezon City

ABSTRACT

Green-lipped mussels (Perna viridis L.) are utilized as bioindicators and bioconcentrators not only for marine radionuclide contamination but also for heavy metal bioaccumulation. Depurated P. viridis were incubated in $^{137}$Cs-spiked aquarium for 4 days. Soluble protein fractions of soft tissue obtained were electrophoresed (SDS-PAGE) to determine the exposure effects of $^{137}$Cs on P. viridis. Results showed the presence of a 154-kDa protein in $^{137}$Cs-spiked samples which could be a potential bioindicator of $^{137}$Cs in P. viridis. Other differences involving two more proteins (~94-kDa and ~61 kDa) are reported.

INTRODUCTION

Artificial radionuclides deposited in the marine ecosystem are due to the global fallout from nuclear weapons testing, discharges from nuclear power facilities and nuclear accidents. The most significant contributors to the total dose ($^{131}$I, $^{134}$Cs and $^{137}$Cs) are systematically considered by various countries (UNSCEAR, 1988). Dose assessments are focused on the contribution of $^{137}$Cs (anthropogenic) and $^{210}$Po (natural), both being considered as the most significant contributors to radiation dose in marine products (Duran et al. 1996).

The beta-gamma emitting $^{137}$Cs is potentially the most harmful isotope in a nuclear fallout due to its long half-life of 30 yrs and high sorption tendency (K') compared with isotopes of Co, Am, Ni, Sr, and I (Penttilae and Kairesalo 1987). These attributes underscore the importance of research into the behavior of radiocesium in the marine food chain (Wang et al. 2000).

Green-lipped mussel (Perna viridis L.), a commercially-valued mollusc, is a known heavy metal bioindicator/bioconcentrator/biomonitor in the environment (Prakash and Rao 1995; Chong and Wang 2000). Yang et al. (1995) reported that these heavy metals, e.g., cadmium, accumulate preferentially in the gills and are bound to metallothioneins.

Little is known on the direct effects of $^{137}$Cs on proteins and gene expression patterns of P. viridis, albeit in the mammalian system, ionizing radiation induces a vast cascade of stress response elements involving tumor suppressors and oncogenes. Among such numerous findings three oncogenes, c-fos, c-jun and c-myc, (so called 'Early Response Genes'), were induced at transcriptional level;
concurrently, and the tumor-suppressor gene \( p53 \) was induced at post-translational level (Matsumoto and Ohnishi 1994).

We report our preliminary findings on the effects of \( ^{137}\text{Cs} \) on the protein profile of green mussels \textit{Perna viridis} (L.) Bivalve: Mytilacea based on the banding patterns of electrophoresed proteins.

**MATERIALS AND METHODS**

**Sampling and Treatment**

\textit{Perna viridis} were collected at Bacoor Bay, Cavite. Depuration was done for 36 hours. Vacuum-filtered seawater were spiked and incubated for 4 days with \( ^{137}\text{Cs} \)-standard (final activity=2.27 dpm/ml). Control set-ups were not spiked.

**Protein Electrophoresis**

Soft tissues were removed and homogenized in phosphate buffered saline, pH 7.6. The homogenate underwent a clearing spin and the crude protein extracts were fractionated in 8% SDS-PAGE and visualized with Coomassie Brilliant Blue (Laemmli 1970).

**RESULTS AND DISCUSSION**

Electrophoretic separation of the soluble protein fractions of \( ^{137}\text{Cs} \)-spiked soft tissues of \textit{Perna viridis} using SDS-PAGE showed the presence of an apparent 154-kDa protein band not found in the control. Two protein bands in the control set-up (~94 kDa and ~61 kDa) were absent in the spiked mussel fraction. Inversely, the spiked sample displayed two bands (~91 kDa and ~59 kDa) absent in the control. The proteins having lower molecular weights (~48 kDa and ~40 kDa) remained constant.

The alterations in the \textit{P. viridis} protein profile could possibly be attributed to response of the mussel to \( ^{137}\text{Cs} \). This unique presence of 154-kDa band could be used as a potential bioindicator for \( ^{137}\text{Cs} \).

The presence of an apparent 154-kDa band could probably be a radioresponse protein produced by \textit{P. viridis} to stress induced by the sublethal toxicity of \( ^{137}\text{Cs} \). The mechanism and capacity for accumulation of another radionuclide, \( ^{226}\text{Ra} \), have been shown in freshwater mussel \textit{V. angasi} (Jeffree 1990). Results on the study on \textit{V. angasi} confirmed that calcium is the model for metabolism of radium by the mussel. \textit{V. angasi} takes up \( ^{226}\text{Ra} \) and stores it in granular deposits as an analogue of calcium. The presence of a similar mechanism in \textit{P. viridis} may exist resulting to changes in protein profile as a result of radioresponse mechanism to stress.

**ACKNOWLEDGMENT**

The work has received support from the following PNRI units: Radiation Protection Unit, Irradiation Services Unit, and Nuclear Training Unit. The authors are thankful for the critical discussions with Mr. Alwyn B. Anfone, Ms. Ma. Teresa Yulo-Nazarea and Dr. Emerenciana B. Duran. The technical assistance of Mr. Dante E. Margate, Mr. Antonio A. Asada, Jr., and Ms. Carol B. Coloma are acknowledged.

**REFERENCES**


Putative Bioindicator of $^{137}$Cs in Perna viridis


