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DOSE ASSESSMENT IN ANABAS TESTUDINEUS AFFECTED FROM EXPOSURE OF WATERBORNE Th-232

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ABSTRACT

Results of the bioaccumulation study and dose assessment of Th-232 in whole-body Anabas testudineus are presented. The objective of this study was to evaluate the effect of Th-232 concentration activity on the laboratory bioaccumulation and total dose rate in Anabas testudineus. Anabas testudineus adults were exposed to different waterborne Th-232 levels: 0 Bq/L (control), 50 Bq/L and 100 Bq/L for 30 day (uptake phase). Whole-body uptakes of Th-232 in Anabas testudineus were calculated and total dose rates using ERICA Assessment Tool were also estimated. The results showed the increase of waterborne Th-232 concentration corresponded to a progressive increase of Th-232 accumulation and total dose rate (internal and external) in whole-body Anabas testudineus. Considering the ERICA dose rate screening value of 10 µGy/h, the findings can be concluded the estimated of total dose rate (0.38 – 1.29 µGy/h) in Anabas testudineus was in order of small magnitude. Nevertheless, these results showed that the Anabas testudineus has a potential to accumulate thorium.

Keywords: *Anabas testudineus*, assessment, bioaccumulation, Th-232, total dose rate

INTRODUCTION

Various contaminants occur in relatively high concentrations in aquatic system which are subjected to receive a radioactive substances, metals and others inputs from industries, agricultural, domestic, accidents, fallout from nuclear weapon testing activities, rare earth processing plant, mining and others. Increased ambient contaminant materials (radionuclides, metals and others) concentration may provoke toxic effects in aquatic organisms which ultimately could lead to a modification of the entire ecosystem structure (Watzin and Roscigno,1997).

Many aquatic organisms such as fish, crustacean and mollusc are an important source of protein among part of the world population. These organisms are capable to accumulate considerable quantities of contaminant materials presented in trace amounts in the ambient water (Iyengar and Narayana Rao,1990). Among the aquatic organisms were used for investigations related to uptake, bioaccumulation and redistribution of both natural and artificial contaminant materials such as fish, clams, green mussels, snail, shrimp, seaweed etc. which particularly are edible types by human (Iyengar, 1983). Variability in the bioaccumulation of contaminants in particular radionuclides from water to aquatic organisms is an important source of uncertainty in generic models for the prediction of radiation doses both to humans and aquatic biota (Monte et al., 2003; Yankovich et al., 2010). The level of radioactive or other contaminants in aquatic biota is commonly defined in terms of a concentration ratio (CR) where:

$$\text{CR (Lkg}^{-1}) = \frac{\text{Activity concentration per kg of the whole-body fresh fish}}{\text{Activity concentration per liter of water}}$$

Many studies on the accumulation of radionuclides in fish have focused on the prediction of CR. The estimation of the fish-water CR may be made from the lab (controlled laboratory experiment) or field measurement (Smith, 2006). It is known that the bioaccumulation of radioactivity in fish is influenced by numerous ecological and environmental factors such as trophic level of the fish species, the length of the food chain, water temperature, water physico-chemistry, fish size and physiology. Some of the experiment factors also believed to influence bioaccumulation includes are the way of experiment carry out and the data generated from field or laboratory studies. Uptake may be via ingestion of contaminated food or direct transfers from the water via the gills (Smith, 2006). Concentration ratios (CRs) are an important component of many radiological dose assessment studies. Typically, these factors are used to support models predicting the activity concentrations of radionuclides in food chains leading to humans and they serve to quantify the health risk associated with ingesting organisms taken from contaminated environments (Twining et al., 1996).

Contaminants such as radionuclides and others reach the aquatic environment as a consequence of human activities, thus aquatic organisms may be exposed to a significant amount of these pollutants. Thorium, Th (including Th-232) is a natural radionuclide present in the aquatic environment. This radionuclide can be found in abundance in aquatic environment such as lakes, ponds, swamps etc. that originated from the mining of other metals, rare earth processing plants and other human activities. Since Th is present in minerals of low solubility, it is generally has been considered insoluble in the water. But it may be adsorbed in particulate and suspended material within water (Langmuir and Herman, 1980). In freshwater environments, Th is relatively unavailable for biological uptake, because it is adsorb strongly to inorganic sediment (Cowart and Burnett, 1994). Most environmental transport of Th is through physical processes where Th adheres to particulate material (inorganic and organic) suspended in water (Langmuir and Herman, 1980).

Anabas testudineus (puyu; climbing perch) is a freshwater fish species commercially grown in Southeast Asian countries (Fig. 1). It is highly demand especially in Malaysia, Indonesia, southern Thailand and the Philippines due to play a major role in providing a protein source for local consumption (Chotipuntu and Avakul, 2010). Their habitats are mostly in rivers, canals, lakes, ponds, swamps and paddy fields (Mijkherjee et al. 2002; Thakur, 2004). *Anabas testudineus* has the habit of migrating from pond to pond. When in water, the fish frequently comes to the surface to breath air. *Anabas testudineus* was selected for this study to assess radiation doses through food chains leading to humans due to the importance of this freshwater fish in the diets for people in Malaysia. Furthermore, its sturdy nature which it can withstand the stress much better than other fishes and; it is ubiquitous and abundant throughout the year in lakes and ponds (Mathew et al., 2013).

Although Th is not biological uptake for aquatic biota (eg. *Anabas testudineus*), however Th may be adsorbed in particulate and suspended material within water and lastly Th which is attached to those materials might be taken up by fish through water intake. On the other hand, Th is probably transformed by cellular processes of metabolism through part of biota body. Thus, these factors can also affect exposure and dose of Th to aquatic biota and such studies will be significant to be performed mainly in the vicinity of the mineral or rare earth processing plant that contains NORM like Th and others naturally occurring radionuclides. Furthermore, currently there is limited study focused on Th-232 exposure in aquatic biota in literature. Therefore, it is important to conduct additional studies regarding the influence of this radionuclide on aquatic biota.The aim of this study was to evaluate the effect of Th-232 concentration activity on the laboratory bioaccumulation and total dose rate in *Anabas testudineus.*

Figure1: Fresh water fish of *Anabas testudineus* (puyu; climbing perch)

MATERIALS AND METHODS

Experiment Materials

Anabas testudineus is a fresh water fish which is ubiquitous and abundant throughout the year in pond, lake, canal and river of Malaysia. For experiment, these healthy fishes were purchased from the ornament fish commercial centre in Bandar Baru Bangi. Natural radionuclide of Th-232 was produced from $Th(NO_3)_4.5H_2O$ salt (Analar grade; purity > 99%; compound of low radioactivity) that purchased from local Sigma-Aldrich supplier. This salt was dissolved in distilled water and the specific activities of Th-232 were measured using Gamma Spectrometry System through its daughter gamma photopeaks of Ac-228 and Tl-208.

Experimental Design

Adult Fishes were acclimated into tap water under experimental conditions (closed-circuit and static aquaria, constantly aerated, light/dark cycle: 10h/14h, pH: 6.5 ± 0.1, temperature: 27 ± 0.1 ^{o}C and salinity: 0) for minimum two weeks prior to commencing the uptake and depuration experiments. They were fed daily with commercial feed pellets in an attempt to ensure good health and no mortality. Twenty individuals of fish with the similar sizes were randomly selected from the acclimatization aquaria and individually exposed to 10 L tap water spiked with 0 Bq/L (control), 50 Bq/L and 100 Bq/L of Th-232 for 30 days in 30 L polypropylene (PP) containers. There was no change in the measured pH of tap water after radionuclide addition. In each experiment, six containers contained 20 individuals of fish for uptake phase were prepared. Experiment waters were renewed and spiked every two days to maintain constant radionuclide concentrations and minimise radionuclide recycling. Experiment waters were aerated to maintain constant levels of dissolved oxygen throughout the experimental exposure period.

During the uptake phase, single selected individuals of fish and water media from each three container were independently sampled at 0,1, 2, 3, 4, 6, 8, 10, 12, 15, 18, 22, 26 and 30 day to represent the sample replications. In this study, the initial concentration of 50 Bq/L was sampled until 40 days to determine an equilibrium concentration. Then, fishes were put in the deep freezer for a while in order to kill them. Fishes were wiped with tissue paper to remove water that affects the fresh weight of fishes. Physical parameter of fishes such as weight, length, height and width were measured and the whole body of each fish was blended individually using food blender or processor. Blended fishes were then placed in 350 mL plastic container and distilled water was added in order to be the same with standard geometry prior to radionuclide activity measurement. About 300 g of water media samples were weighed and placed in 350 mL plastic container for counting. These two types of blended whole-body fish and water media samples were counted using gamma spectrometry system for 28800 and 3600 seconds, respectively. The activity concentrations for each type of sample were averaged from the triplicates of the samples. Concentration ratios of Th-232 were then calculated as explain in introduction part and whole-body total dose rates were estimated using ERICA Assessment Tool.

RESULTS AND DISSCUSSION

Activity Concentration of Th-232 in Whole-body *Anabas testudineus*

Activity concentrations of Th-232 in whole-body *Anabas testudineus* after exposed to different levels of this radionuclide for 30 days through water media are presented in Fig. 2. The results found that the Th-232 activity concentrations in respectively were differed and varied between 4.31 ± 1.51 Bq/kg fw. to 8.90 ± 3.12 Bq/kg fw. (control; 0 Bq/L), 11.40 ± 3.42 Bq/kg fw. to 44.32 ± 15.52 Bq/kg fw. (50 Bq/L exposure) and 34.96 ± 17.83 Bq/kg fw. to 56.05 ± 28.31 Bq/kg fw. (100 Bq/L exposure). This finding was proved by a one way ANOVA analysis that the activity concentration of Th-232 in *Anabas testudineus* at different exposure concentration have significant difference with $p = 0.000$. This indicating the accumulation of Th-232 in *Anabas testudineus* was depending to exposure level of radionuclide. On the other hand, the increase Th-232 concentration in the water corresponded to a progressive increase of Th-232 in whole-body *Anabas testudineus* in particular for exposure concentration of 100 Bq/L. This result can be related to physiological differences in uptake and excretory mechanism as well as health condition for each fish. Thus, these factors might be affected on the uptake of Th-232 by *Anabas testudineus* that can be observed a drastic increase of Th-232 at day 2 experiments for 100 Bq/L.

Meanwhile, the different uptake trends between 50 Bq/L and 100 Bq/L where the uptake 100 Bq/L of Th-232 by *Anabas testudineus* was slow compared to 50 Bq/L. This due to Th-232 is relatively radiotoxic and unavailable for biological uptake, thus excretory kinetic might be played important role to release faster a radiotoxic of Th-232 from the whole-body *Anabas testudineus*. This can be concluded that exposure to different Th-232 activity concentrations were significantly alter the accumulation of Th-232 in *Anabas testudineus.* These differences or variations also might be due to other reasons such as metabolisms, excretory patterns, longevity, types of exposures etc.

Uptake Kinetic of Th-232 in Whole-body *Anabas testudineus*

The 30 days (and 40 days for 50 Bq/L) uptake kinetics of Th-232 in whole-body *Anabas testudineus* at different waterborne exposure concentrations are illustrated in Fig. 3. Uptake of Th-232 in whole-body *Anabas testudineus* displayed exponential kinetics at all of the two exposure concentrations. These observed that exponential kinetic models tended to reach a steady state (saturation) after 15 days (50 Bq/L exposure) and 4 days (100 Bq/L exposure) i.e. faster than the experiment condition of 30 days. This curve shapes indicated that equilibrium concentration ratios were approximately reached at 0.95 and 0.67 respectively for 50 Bq/L and 100 Bq/L exposures. Thus, results showed the bioconcentration of Th-232 in whole-body *Anabas testudineus* was directly proportional to the Th-232 concentration in water media.

Meanwhile, statistical analysis indicated that mean uptake rate in *Anabas testudineus* were not differ significantly at all exposure concentrations with the range of 0.10 – 0.13 L/kg.d and it was comparatively constant throughout the 30 days depuration period (Table 1 and 2). This indicated the uptake rates of Th-232 by whole-body *Anabas testudineus* were not depend on waterborne or water media concentrations. According to previous researcher that variation of contaminants accumulate or uptake by organisms depends on their size-dependent feeding and breath behaviour, surface area to volume ratios, as well as concentrations of enzymes may also play a role in influencing the contaminant uptake. Furthermore, physiological differences in metabolic demand were also the primary cause for the differential contaminant accumulation (Bruner et al., 1994). These statement is also support our finding that our fish samples are not all the same size and their different of surface area to volume ratios which could be affected the accumulation or uptake of Th-232 by *Anabas testudineus*. Besides that, the healthy condition and physiology of fish also influenced the metabolism or mechanism of uptake or excretory contaminants like radiotoxic of Th-232. Thus, these reasons are among the factors that differentiated Th-232 accumulation in *Anabas testudineus*.

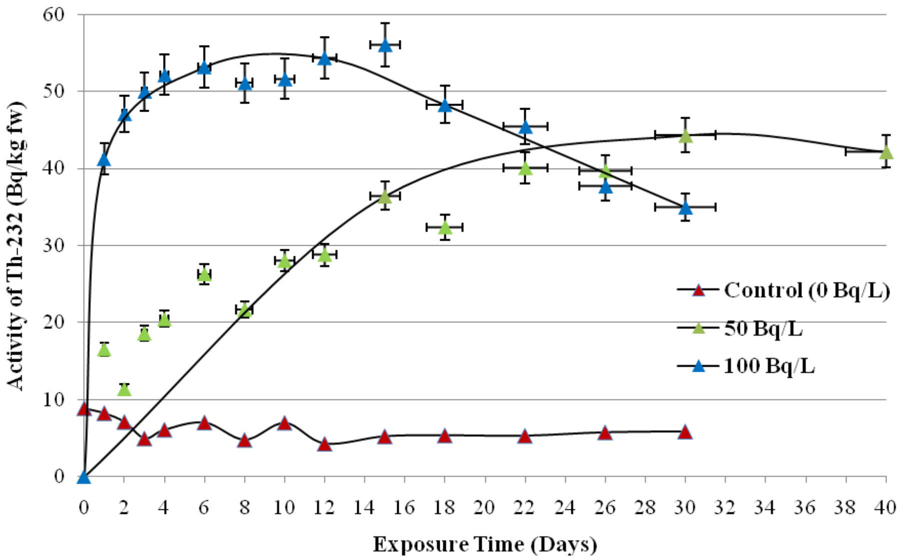

Figure 2: Activity concentration of Th-232 in whole-body *Anabas testudineus* during 30 days uptake experiment

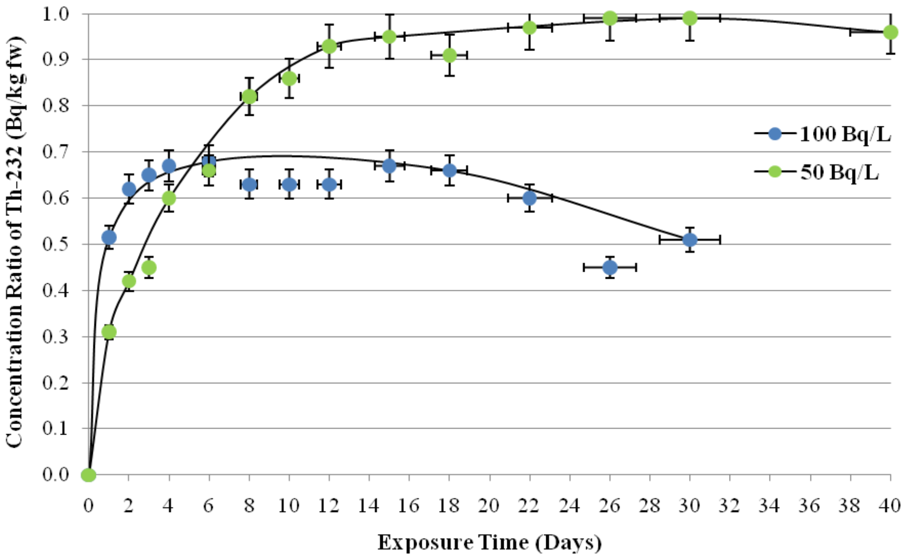

Figure 3: Whole-body uptake kinetic of Th-232 in *Anabas testudineus* from different waterborne exposure concentration

Total Dose Rate Estimated Using the ERICA Assessment Tool

The total dose rates and estimated from measured Th-232 activity in whole-body *Anabas testudineus* at different exposure concentration for the time period of 30 days are shown in Table 1 and 2. These data were analysed using the ERICA Assessment Tool based on the actual data measured in this study. The total dose rates for all *Anabas testudineus* were much lower and vary from 0.38 – 1.02 µGy/h (50 Bq/L exposure) and 0.81 – 1.29 µGy/h (100 Bq/L exposure). These indicated that the higher waterborne activity concentrations of Th-232 were directly reflected in higher total dose rate to the *Anabas testudineus*. Nonetheless, those estimated total dose rates in *Anabas testudineus* were in order of small magnitude compared to the ERICA dose rate screening value (10 µGy/h).

Plan for the Future Study

Due to time constraints and limited laboratory space, only one species of *Anabas testudineus* for freshwater fish was focused in this study. To compare the accumulation and to estimate the radiological dose and risk of Th-232 to freshwater fish, others fish species will also be studied in the future.

Table 1: Uptake rate and total dose rate at 50 Bq/L exposure concentrations generated from the ERICA Assessment Tool

Day	**Activity of Th-232**		**Concentration Ratio (CR) (L/kg)**	**Uptake Rate (L/kg.d)**	**Fish Mass (kg)**	**High (m)**	**Width (m)**	**Length (m)**	**Total Dose Rate per Organism (µGy/h)**	**Risk Quotient (Expected Value) (Unitless)**	**Risk Quotient (Conservative Value) (Unitless)**
	Water (Bq/L)	**Fish (Bq/kg fw.)**									
0	55.44 ± 15.52	0.00	0.00	0.00	0.038	0.035	0.026	0.111	0.000	0.000	0.000
1	52.10 ± 15.63	16.56 ± 6.46	0.31	0.31	0.030	0.032	0.018	0.107	0.382	0.038	0.115
2	28.67 ± 14.34	11.40 ± 3.42	0.42	0.21	0.039	0.032	0.021	0.110	0.263	0.026	0.079
3	41.49 ± 18.67	18.61 ± 6.51	0.45	0.15	0.036	0.029	0.020	0.115	0.429	0.043	0.129
4	34.14 ± 10.58	20.47 ± 7.78	0.60	0.15	0.033	0.029	0.017	0.117	0.472	0.047	0.142
6	40.25 ± 18.11	26.29 ± 9.46	0.66	0.11	0.039	0.032	0.020	0.120	0.606	0.061	0.182
8	26.64 ± 10.12	21.71 ± 7.38	0.82	0.10	0.020	0.026	0.015	0.093	0.501	0.050	0.150
10	36.39 ± 13.38	28.03 ± 9.53	0.86	0.09	0.031	0.031	0.020	0.108	0.646	0.064	0.194
12	30.43 ± 10.78	28.80 ± 10.08	0.93	0.08	0.036	0.029	0.019	0.118	0.664	0.066	0.199
15	38.34 ± 10.25	36.42 ± 12.60	0.95	0.06	0.018	0.024	0.014	0.088	0.840	0.084	0.252
18	36.12 ± 12.25	32.38 ± 9.71	0.91	0.05	0.026	0.025	0.016	0.107	0.747	0.075	0.224
22	41.14 ± 16.46	40.04 ± 14.41	0.97	0.04	0.022	0.025	0.015	0.099	0.923	0.092	0.277
26	40.41 ± 18.18	39.67 ± 13.89	0.99	0.04	0.024	0.027	0.016	0.100	0.915	0.092	0.274
30	44.73 ± 18.13	44.32 ± 15.52	0.99	0.03	0.025	0.028	0.018	0.102	1.020	0.102	0.307
40	43.98 ± 13.99	42.20 ± 14.77	0.96	0.02	0.044	0.029	0.021	0.114	0.973	0.097	0.292

Table 2: Uptake rate and total dose rate at 100 Bq/L exposure concentrations generated from the ERICA Assessment Tool

	Activity of Th-232										
Day	**Water (Bq/L)**	**Fish (Bq/kg fw.)**	**Concentration Ratio (CR) (L/kg)**	**Uptake Rate (L/kg.d)**	**Fish Mass (kg)**	**High (m)**	**Width (m)**	**Length (m)**	**Total Dose Rate per Organism (μGy/h)**	**Risk Quotient (Expected Value) (Unitless)**	**Risk Quotient (Conservative Value) (Unitless)**
0	83.33 ± 34.16	0.00	0.00	0.00	0.019	0.029	0.015	0.084	0.000	0.000	0.000
1	80.08 ± 31.23	41.25 ± 17.45	0.52	0.52	0.016	0.024	0.013	0.088	0.951	0.095	0.285
2	75.95 ± 26.58	47.09 ± 23.53	0.62	0.31	0.012	0.021	0.011	0.075	1.090	0.109	0.326
3	76.92 ± 26.15	49.99 ± 25.50	0.65	0.22	0.015	0.022	0.013	0.081	1.150	0.115	0.346
4	77.81 ± 27.23	52.13 ± 25.54	0.67	0.17	0.015	0.026	0.014	0.078	1.200	0.120	0.361
6	78.19 ± 27.05	53.17 ± 26.05	0.68	0.11	0.020	0.028	0.016	0.082	1.230	0.123	0.368
8	81.11 ± 30.01	51.10 ± 27.08	0.63	0.08	0.016	0.026	0.013	0.083	1.180	0.118	0.353
10	81.94 ± 28.35	51.62 ± 27.36	0.63	0.06	0.015	0.023	0.013	0.079	1.190	0.119	0.357
12	86.27 ± 30.19	54.35 ± 27.23	0.63	0.05	0.015	0.025	0.014	0.083	1.250	0.125	0.376
15	83.66 ± 29.03	56.05 ± 28.31	0.67	0.04	0.014	0.023	0.013	0.079	1.290	0.129	0.388
18	73.12 ± 25.67	48.26 ± 27.03	0.66	0.04	0.016	0.026	0.014	0.080	1.110	0.111	0.334
22	75.75 ± 26.59	45.45 ± 23.13	0.60	0.03	0.022	0.026	0.016	0.095	1.050	0.105	0.314
26	83.80 ± 29.33	37.71 ± 18.59	0.45	0.02	0.016	0.027	0.014	0.078	0.869	0.087	0.261
30	68.53 ± 23.51	34.96 ± 17.83	0.51	0.02	0.025	0.028	0.018	0.102	0.806	0.081	0.242

CONCLUSIONS

Generated data from short-time laboratory experiments showed that presence of Th-232 in different waterborne concentration significantly influenced the bioaccumulation, depuration and total dose rate in freshwater fish of *Anabas testudineus*. Generally, the results displayed the increase of waterborne Th-232 concentration corresponded to a progressive increase of Th-232 accumulation and total dose rate (internal and external) in whole-body *Anabas testudineus*. Considering the ERICA dose rate screening value of 10 µGy/h, the findings can be concluded the estimated of total dose rate (0.38 – 1.29 µGy/h) in *Anabas testudineus* was in order of small magnitude. Total dose rate received by *Anabas testudineus* estimated using ERICA Assessment Tool showed that this freshwater fish not contaminated with both (50 Bq/L and 100 Bq/L) exposure concentration of Th-232. Nonetheless, in this study might ideally address the kinetics of uptake from this *Anabas testudineus* to consider this fresh water fish has a potential to accumulate thorium.

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REFERENCES

Bruner, K.A. Fisher, S.W. and Landrum, P.F. (1994). The role of the zebra mussel, dreissena polymorpha, in contaminant cycling: I. the effect of body size and lipid content on the bioconcentration of PCBs and PAHs, *J. Great Lakes Res.* 20(4): 725-734.

Chotipuntu, P. and Avakul, P. (2010). Aquaculture potential of climbing perch, Anabas Testudineus in brackish water Walailak, *J. Sci. & Technol.* 7(1): 15-21.

Cowart, J.B. and Burnett, W.C. (1994). The distribution of uranium and thorium decay series radionuclides in the environment – a review, *J. Environ. Quality*. 23: 651-662.

Iyengar, M.A.R. (1983). Studies on the distribution of natural radionuclides in the marine organisms. Doctoral thesis, University of Bombay.

Iyengar, M.A.R. and Narayana Rao, K. (1990). The environmental behaviour of radium, IAEA Tech. Rep. Series No. 310, Vienna, Austria. 467-684.

Langmuir, D. and Herman, J.S. (1980). The mobility of thorium in natural waters at low temperatures, *Geochim. Cosmochim. Acta.* 44: 1753-1766.

Mathew, E. Sunitha, P.T. and Thomas, P.L. (2013). Effect of different concentrations of detergent on dissolved oxygen consumption in *Anabas testudineus*, *IOSR J. Environ. Sci. Toxicol. Food Technol.(IOSR-JESTFT).* 5(3): 2319-2399.
Mijkherjee, M. Praharaj, A. and Das, S. (2002). Conservation of endangered fish stocks through artificial propagation and larval rearing technique in West Bengal, India, *Aquatic Asia*. 7(2): 8-11.

Monte, L., Brittain, J.E., Hakanson, L., Heling, R., Smith, J.T. and Zheleznyak, M. (2003). Review and assessment of models used to predict the fate of radionuclides in lakes, *J. Environ. Radioact.* 69: 177-205.

Smith, J.T. (2006). Modelling the dispersion of radionuclides following short duration releases to rivers: Part 2. Uptake by fish, *Sc. Tot. Environ*. 368: 502-508.

Thakur, D.P. (2004). New species studied for aquaculture potential by aquaculture CRSP researchers, *Aquanews.* 19(1): 1-6.

Twining, J.R. Ferris, J.M. and Markich, M.J. (1996). Bioaccumulation of 137Cs and 85Sr by an Australian sub-tropical freshwater toleost (Bidyanus bidyanus), *Sci. Tot. Environ*. 192: 245-257.

Yankovich, T.L., Batlle, J.V.I., Vives-Lynch, S., Beresford, N.A., Barnett, C.L., Beaugelin-Seiller, K., Brown, J.E., Cheng, J.-J., Copplestone, D., Heling, R., Hosseini, A., Howard, B.J., Kamboj, S., Kryshev, A.I., Nedveckaite, T., Smith, J.T. and Wood, M.D. (2010). An international model validation exercise on radionuclide transfer and doses to freshwater biota, *J. Radiol. Protect*. 30: 299-340.

Watzin, M.C. and Roscigno, P.R. (1997). The effects of zinc contamination on the recruitment and early survival of benthic invertebrates in an estuary, *Mar. Pollut. Bul*. 34: 443-455.