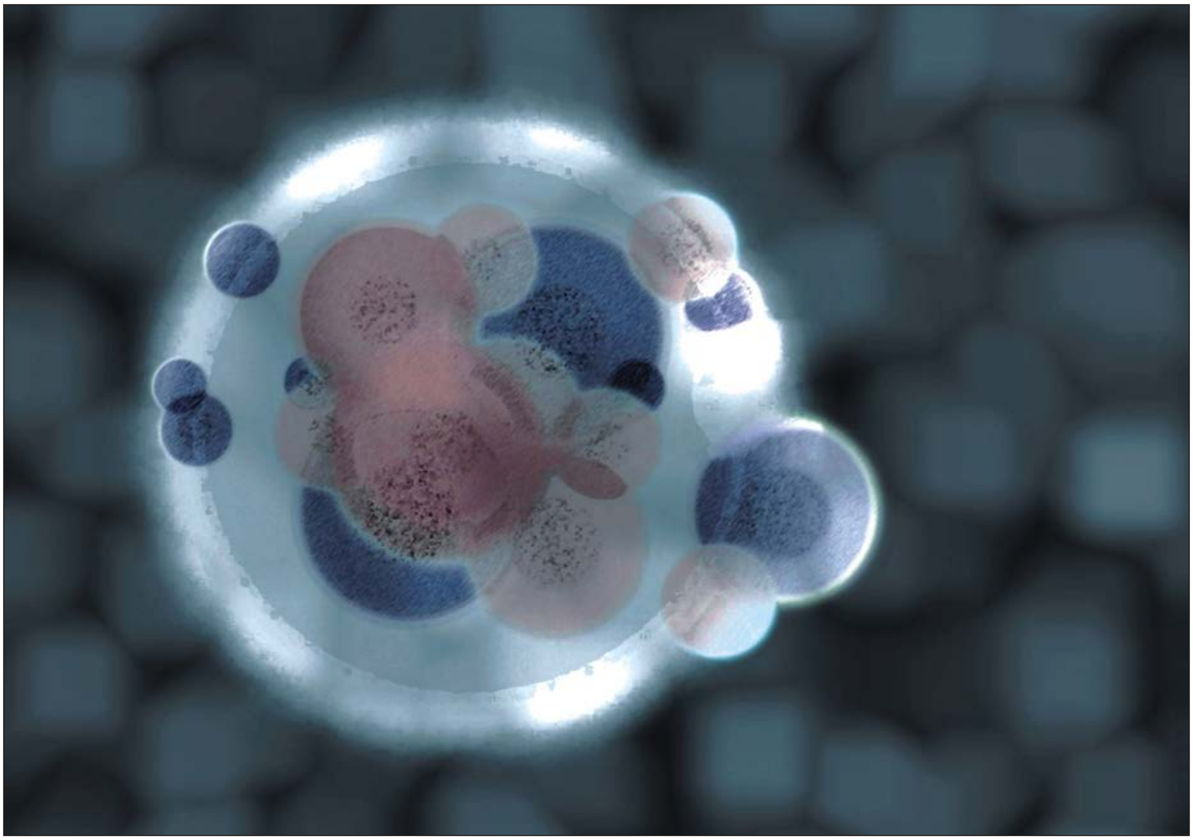

Journal of Biological Sciences

ISSN 1727-3048

Journal of Biological Sciences 8 (3): 644-648, 2008
ISSN 1727-3048

Feasibility of Producing Selenium-Enriched Water Lettuce (*Pistia stratiotes* L.)

[1]Anut Chantiratikul, [2]Panida Atiwetin and [3]Piyanete Chantiratikul
[1]Faculty of Veterinary Medicine and Animal Science, Mahasarakham University,
Muang, Maha Sarakham, 44000, Thailand
[2]Department of Agricultural Production Technology, Faculty of Technology,
Mahasarakham University, Muang, Maha Sarakham, 44000, Thailand
[3]Department of Chemistry, Faculty of Science, Mahasarakham University,
Kantarawichai, Maha Sarakham, 44150, Thailand

Abstract: The feasibility of producing selenium-enriched water lettuce (*Pistia stratiotes* L.) was studied by cultivating water lettuce in Hoagland's solution containing 0, 20, 40, 60 and 80 mg Se from sodium selenite/L. There were 4 replicates in each Se concentration. Each replicate consisted of 30 plants of water lettuce. Three plants of water lettuce in each replicate were sampled on day 0, 1, 2, 3 and 4 of the experiment. The samples were washed with deionized water, separated for leaves and roots and finally dried at 65°C. Prior to Se determination, leaf or root samples were pooled by replicate. The finding revealed that Se concentrations in leaves and roots of water lettuce increased significantly ($p<0.05$) with increasing Se concentration in Hoagland's solution and day of cultivation. However, Se concentration in leaves was lower than that of roots. Water lettuce cultivated in the solution containing 60 to 80 mg Se L^{-1} exhibited the yellow leaves and died in day 2 and 3. Therefore, the appropriate Se concentration and duration for producing Se-enriched water lettuce were 20 to 40 mg Se L^{-1} and 2 to 3 days of cultivation. The leaves of water lettuce cultivated in those conditions contained 11.14-13.50 and 21.06-29.55 mg Se kg^{-1}, respectively.

Key words: Water lettuce, *Pistia stratiotes* L., selenium enrichment

---

INTRODUCTION

Selenium (Se) is a crucial trace element in human and animal nutrition (McDowell, 1992; Underwood and Suttle, 1999). It is an essential component of a number of enzymes like glutathione peroxidase (Arthur, 1997) and plays an important role in thyroid metabolism (Köhrle *et al*., 2005). Furthermore, daily Se supplementation in amounts up to 200 μg can reduce colorectal and prostate cancer (Schrauzer, 2002; Kim and Mahan, 2003). An insufficient Se intake has been linked with impaired thyroid metabolism, infertility, reduced response to vital infection and in more serious situations, cardio-and skeletal-myopathies (MacRae, 2005). Therefore, numerous studies have been concentrated on increasing Se content in animal products to improve Se status of the consumers. The outcomes of those studies revealed that organic Se in the form of Se-enriched yeast significantly increased ($p<0.05$) Se contents in meat of broilers (Olivera *et al*., 2005), pork (Mahan and Parrett, 1996; Zhan *et al*., 2007), milk (Ortman and Pehrson, 1999; Knowles *et al*., 1999; Juniper *et al*., 2006; Pechova *et al*., 2008) and eggs (Payne *et al*., 2005; Skrivan *et al*., 2006; Pan *et al*., 2007) when compared with inorganic Se in the form of selenite. However, Se-enriched yeast is an expensive commercial product and its production process requires complex and high technology (Suhajda *et al*., 2000). On the other hand, the production of Se-enriched plants is more practical (Sugihara *et al*., 2004; Tsuneyoshi *et al*., 2006). Thus, the Se-enriched plants have been extensively reported in prospect of human nutrition in broccoli sprout (Finley *et al*., 2001), green onions (*Allium fistulosum*) (Kapolna and Fodor, 2006), garlic (Tsuneyoshi *et al*., 2006) and sprouts of several plants (Lintschinger *et al*., 2000; Sugihara *et al*., 2004). Jiakui and Xiaolong (2004) produced Se-enriched malt and fed it to laying hens. Although, they found that Se from sodium selenite and Se-enriched malt insignificantly deposited in the eggs, the result showed that Se-enriched plant could be used as Se source in animal diets.

Water lettuce (*Pistia stratiotes* L.) is a free-floating freshwater macrophyte and known as one of the most important tropical aquatic weeds (Labrada and Fornasari, 2002). It presents a high growth rate and has been successfully used for the removal of toxic metals such as mercury (Hg), cadmium (Cd), chromium (Cr), cupper (Cu),

---

Corresponding Author: Anut Chantiratikul, Faculty of Veterinary Medicine and Animal Science, Mahasarakham University, Muang, Maha Sarakham, 44000, Thailand Tel: +66-87-173-8777 Fax: +66-43-742-823

zinc (Zn) and manganese (Mn) from pollutant water (De *et al*., 1985; Sridhar, 1986; Satyakala and Jamil, 1992; Miretzky *et al*., 2004; Skinner *et al*., 2007). The above reports exhibited that water lettuce has high capacity in metal absorption. Therefore, water lettuce can be possibly used to produce Se-enriched plant for animal nutrition. The objective of this study was to determine the effect of Se concentrations on Se accumulation in water lettuce.

MATERIALS AND METHODS

The experiment was conducted in October to November, 2007. Water lettuce plants were corrected from a natural pond in Maha Sarakham province located in northeastern Thailand. The collected plants were washed thoroughly and maintained with tap water in plastic containers for 5 days. Subsequently, thirty of the same size plants with an approximate diameter of 5 cm were selected and introduced into each plastic container (30×40×8 cm) containing 0, 20, 40, 60 and 80 mg Se L^{-1} in Hoagland's solution. Sodium selenite (Fluka, Germany) was used as the Se source. There were four replicates in each Se concentration. Three plants of water lettuce were sampled periodically at day 0, 1, 2, 3 and 4 of the experiment from each replicate. The sampled plants were washed thoroughly with deionized water, separated into leaves and roots and later dried at 65°C for 48 h. Dried individual sample of leaves and roots was ground through 1 mm screen and separately pooled by replicate before determination of Se concentration. Approximately 0.5 g of pooled samples of leaves and roots were digested in a mixture of 1 mL HNO_3 and 9 mL deionized water until the solution was cleared. The mineralisates were diluted with deionized water to the final volume of 25 mL. Selenium was determined by inductively coupled plasma mass spectrometer (ICP-MS model Elan-e, Perkin-Elmer SCIEX, USA) according to Joaquim *et al*. (1997).

Statistical analysis: The data of Se concentrations in leaves and roots of water lettuce were in completely randomize design using the general linear model procedure (SAS, 1996). The differences among means of Se concentrations in leaves or roots were compared by Duncan's New Multiple Range Test (Steel and Torries, 1989). A probability level of $p<0.05$ was considered to be statistically significant.

RESULTS AND DISCUSSION

The results demonstrated that Se concentrations in leaves of water lettuces increased significantly ($p<0.05$) with increasing Se concentration in the solution. The Se content in leaves of water lettuce cultivated in solution containing 0 mg Se L^{-1} ranged from 0.25 to 0.48 mg L^{-1} (Table 1). However, Se concentrations in leaves of water lettuces cultivated in solutions containing 20 and 40 mg Se L^{-1} increased significantly ($p<0.05$) from day 0 to 3 and seemed to be gradually declined ($p>0.05$) thereafter. Selenium concentrations in leaves of water lettuces cultivated in solution containing 60 mg Se L^{-1} increased significantly ($p<0.05$) from day 0 to 2, but decreased abruptly ($p<0.05$) in day 3.

The Se concentrations in roots of water lettuces increase significantly ($p<0.05$) with increasing Se concentration in the solution and exposure period (Table 2). Selenium deposition in roots was higher than that in leaves of water lettuce (Table 1). Unfortunately, there was no available scientific information of Se

Table 1: Selenium concentration (mg kg^{-1}) in leaves of water lettuce cultivated in the solution containing different Se concentrations

Day of cultivation	Se concentrations (mg L^{-1}) 0	20	40	60	80	SEM
0	0.48^{ae}	0.09^{ch}	0.17^{bh}	0.07^{ch}	0.09^{cf}	0.04
1	0.30^{dfg}	5.30^{cg}	12.55^{bg}	14.85^{bg}	19.76^{ae}	1.62
2	0.25^{dg}	11.14^{cf}	21.06^{bf}	43.04^{ae}	#	4.04
3	0.35^{cef}	13.50^{be}	29.55^{ae}	31.60^{af}	#	3.61
4	0.33^{cf}	13.17^{be}	26.46^{ae}	#	#	12.12

^{abcd}: Values in row with different superscripts are significantly different at $p<0.05$, ^{efgh}: Values in column with different superscripts are significantly different at $p<0.05$, #: Water lettuce died

Table 2: Selenium concentration (mg kg^{-1}) in roots of water lettuce cultivated in the solution containing different Se concentrations

Day of cultivation	Se concentrations (mg L^{-1}) 0	20	40	60	80	SEM
0	0.63^{bcg}	0.96^{af}	0.87^{abh}	1.06^{ah}	0.41^{cf}	0.08
1	1.25^{dg}	55.81^{ce}	81.08^{bg}	102.61^{bf}	130.24^{ae}	10.69
2	3.13^{cf}	56.51^{ce}	169.43^{bef}	313.62^{ae}	#	42.20
3	5.48^{ce}	77.34^{bce}	162.48^{bf}	353.71^{ae}	#	44.25
4	1.12^{cg}	94.60^{be}	232.66^{ae}	#	#	109.46

^{abcd}: Values in row with different superscripts are significantly different at $p<0.05$, ^{efgh}: Values in column with different superscripts are significantly different at $p<0.05$, #: Water lettuce died deposition in water lettuce. However, the previous studies similarly observed Cr and Hg accumulations in roots of water lettuce was 3 to 5-folds (Satyakala and Jamil, 1992) and 4-folds (De *et al.*, 1985) higher than that in leaves, respectively. Additionally, Fargasova (2004) reported Se accumulation was higher in the roots than in the cotyledon of *Sinapis alba* L. Roots of aquatic plants naturally submerged in the water. It was therefore the first part of plant to accumulate minerals (Skinner *et al.*, 2007). The translocation of metals from roots to shoots generally was very low. Xiong (1998) observed about 90% of lead (Pb) taken up remained in the underground parts of plants. Similarly, Fargasova (2004) mentioned that translocation of Se to aboveground plant parts was probably slow. Because Se is transported predominantly in the xylem and presumably the greater leaf surface area contributed to a relatively higher respiration rate and increased movement of Se to the transpiring leaves of plants (Banuelos *et al.*, 1997).

Selenium accumulation in water lettuce increased rapidly from the first day of cultivation. Selenium concentrations in leaves and roots on day 1 increased 58.88 to 219.55-folds and 58.13 to 327.65-folds, respectively, as compared with those on day 0. However, Se concentrations in leaves and roots increased (0.73 to 3.06-folds) in a declining rate from day 2 to 4. The above results indicated that water lettuce greatly absorbed and accumulated Se in the first 24 h of cultivation. The foregoing findings similarly found that higher than 85% of Hg (De *et al.*, 1985), Pb, Cr, Mn and Zn (Miretzky *et al.*, 2004) in the water were absorbed by water lettuce within 24 h. The rate of the metal uptake process was dependent on the metal concentration in plants (Miretzky *et al.*, 2004). Thus, the decreasing rate of Se absorption was probably due to higher Se concentration in water lettuce and the decreased mineral concentration in the water (Attionu, 1976).

Toxic metal accumulation produced significant physiological and biochemical responses (Phalson, 1989). Water lettuce cultivated in solution containing 60 and 80 mg Se L^{-1} showed partly yellowing of leaves and died within 3 and 2 days, respectively. However, water lettuce showed yellowing of leaves in day 4 when it was cultivated in the solution containing 40 mg Se L^{-1}. The yellowing of leaves was previously found in water lettuce cultivated in the solution containing 20 mg L^{-1} of Hg (De *et al.*, 1985) and 25 to 50 mg L^{-1} of Cr (Satyakala and Jamil, 1992). Furthermore, the roots of the dead water lettuce appeared to reduce in length. An excess of Se in plant can adversely affect chlorophyll content (Fargasova, 2004) and morphology of roots (Hartikainen *et al.*, 2001; Fargasova, 2004). Se toxicity inhibited severely the production of chlorophyll (Fargasova, 2004). Subsequently, the yellowing of leaves or chrolosis in water lettuce occurred in the present study. The restricted root elongation of water lettuce might be caused by the damage of the root plasma membrane during Se penetration into the root cells (Fargasova, 2004). The obtained results demonstrated that water lettuce could tolerate Se concentration approximately 20 to 40 mg L^{-1}.

Absorbed Se will be converted metabolically in chloroplast to selenoprotein, predominantly in the form of selenomethionine, which is a component of protein in tissues of plants (Leustek and Saito, 1999; Tinggi, 2003). Thus, the suitable plants for Se-enriched plant production should contain high protein, absorb and accumulate Se markedly and convert effectively inorganic Se to organic Se. Water lettuce contained 15 to 35% of protein, mostly in leaves (Rao and Reddy, 1984; Henry-Silva and Camargo, 2006). It is possibly used for Se-enriched production. The current results of Se accumulation, Se tolerant concentration and duration indicated that the appropriate Se concentration and duration for producing Se-enriched water lettuce were 20 to 40 mg Se L^{-1} and 2 to 3 days, respectively. Selenium concentrations in leaves of water lettuce cultivated in the solution containing 20 and 40 mg Se L^{-1} were 11.14 to 13.50 and 21.06 to 29.55 mg kg^{-1}, respectively. The productions of Se-enriched plants have been extensively studied in several edible plants. Kapolna and Fodor (2006) reported total Se concentration in green onions (*Allium fistulosum*) reached the 61.8 mg kg^{-1} level when applying selenite at a concentration of 100 mg L^{-1} as Se source. Finley *et al.* (2001) produced high-Se broccoli sprout contained 62.3 mg Se kg^{-1} using selenate at a concentration of 25 mg L^{-1} as Se source. The Se concentration of Se-enriched malt was 60.5 mg kg^{-1} when treated with water containing 90 mg Se L^{-1} of selenite (Jiakui and Xiaolong, 2004). The Se concentrations in Se-enriched plants previously reported were higher than that found in water lettuce in this study. Therefore, further study for producing Se-enriched water lettuce should focus on factors affecting Se absorption and accumulation such as levels and sources of Se, age of water lettuce and the efficiency of conversion of inorganic Se to organic Se.

CONCLUSIONS

Selenium concentrations in leaves and roots of water lettuce increased significantly ($p<0.05$) with increasing Se concentration in the solution and exposure period. The suitable Se level and duration for producing Se-enriched water lettuce were 20 to 40 mg Se L^{-1} and 2 to 3 days, respectively. Selenium concentrations in leaves of water lettuce cultivated in those conditions contained 11.14 to 29.55 mg Se kg^{-1}.

ACKNOWLEDGMENTS

This research (research project No. 5001103) was financially supported by Thai Government budget fiscal year 2007 and Mahasarakham University. The authors wish to thank Witpol Thosaikham, Phatarada Suthamwong and Rinnapa Sumsanuk for laboratory and technical assistance.

REFERENCES

Arthur, J.R., 1997. Non-Glutathione Peroxidase Functions of Selenium. In: Biotechnology in the Feed Industry, Lyons, T.P. and K.A. Jacques (Eds.). Redwood Books, Wiltshire, England, pp: 143-154.

Attionu, R.H., 1976. Some effects of water lettuce (*Pistia stratiotes*, L.) on its habitat. Hydrobiologia, 50 (3): 245-254.

Banuelos, G.S., H.A. Ajwa, L. Wu, X. Guo, S. Akohoue ans S. Zambrzuski, 1997. Selenium-induced growth reduction in *Brassica* land races considered for phytoreduction. Ecotoxicol. Environ. Safety, 36 (3): 282-287.

De, A.K., A.K. Sen and D.P. Modak, 1985. Studies on toxic effects on Hg(II) on *Pistia* stratiotes. Water Air Soil Pollut., 24 (4): 351-360.

Fargasova, A., 2004. Toxicity comparison of some possible toxic metals (Cd, Cu, Pb, Se, Zn) on young seedling of *Sinapis alba* L. Plant Soil Environ., 50 (1): 33-38.

Finley, J.W., C. Ip, D.J. Lisk, C.D. Davis, K.J. Hintze and P.D. Whanger, 2001. Cancer-protective properties of high-selenium broccoli. J. Agric. Food Chem., 49 (5): 2679-2683.

Hartikainen, H., L. Pietola, A. Simojoki and T. Xue, 2001. Quantification of fine root responses to selenium toxicity. Agric. Food Sci. Finland, 10 (1): 53-58.

Henry-Silva, G.G. and A.F.M. Camargo, 2006. Chemical composition of floating aquatic macrophytes used to treat of aquaculture wastewater. Planta Daninha, 24 (1): 21-28.

Jiakui, L. and W. Xiaolong, 2004. Effect of dietary organic versus inorganic selenium in laying hens on the productivity, selenium distribution in egg and selenium content in blood, liver and kidney. J. Trace Elem. Med. Biol., 18 (1): 65-68.

Joaquim, A.N., Y. Gelinas, A. Krushevska and R.M. Barnes, 1997. Determination of elements in biological and botanical materials by inductively coupled plasma atomic emission and mass spectrometry after extraction with a tertiary amine reagent. J. Anal. At. Spectrom., 12 (10): 1239-1242.

Juniper, D.T., R.H. Phipps, A.K. Jones and G. Bertin, 2006. Selenium supplementation of lactating dairy cows: Effect on selenium concentration in blood, milk, urine and feces. J. Dairy Sci., 89 (9): 3544-3551.

Kapolna, E. and P. Fodor, 2006. Speciation analysis of selenium enriched green onions (*Allium fistulosum*) by HPLC-ICP-MS. Microchem. J., 84 (1-2): 56-62.

Kim, Y.Y. and D.C. Mahan, 2003. Biological aspects of selenium in farm animals. Asian-Aust. J. Anim. Sci., 16 (3): 435-444.

Knowles, S.O., N.D. Grace, K. Wurms and J. Lee, 1999. Significance of amount and form of dietary selenium on blood, milk and casein selenium concentrations in grazing cows. J. Dairy Sci., 82 (2): 429-437.

Köhrle, J., F. Jakob, B. Contempre and J.E. Dumont, 2005. Selenium, the thyroid and the endocrine system. Endocrine Rev., 26 (7): 944-984.

Labrada, R. and L. Fornasari, 2002. Management of the Worst Aquatic Weeds in Africa. FAO Efforts and Achievements During the period 1991-2001. FAO, Rome.

Leustek, T. and K. Saito, 1999. Sulfate transport and assimilation in plants. Plant Physiol., 120 (3): 637-643.

Lintschinger, L., N. Fuchs, J. Moser, D. Kuehnelt and W. Goessler, 2000. Selenium-enriched sprouts. A raw material for fortified cereal-based diets. J. Agric. Food Chem., 48 (11): 5362-5368.

MacRae, J.C., 2005. Animal products for a healthy diet. Proceedings of Integrating Livestock-Crop Systems to Meet the Challenges of Globalization. Vol. 1. Khon Kaen, Thailand, pp: 179-188.

Mahan, D.C. and N.A. Parrett, 1996. Evaluating the efficacy of selenium-enriched yeast and sodium selenite on tissue selenium retention and serum glutathione peroxidase activity in grower and finisher swine. J. Anim. Sci., 74 (12): 2967-2974.

McDowell, L.R., 1992. Minerals in Animal and Human Nutrition. Academic Press, Inc. San Diego, California.

Miretzky, P., A. Saralegui and A.F. Cirelli, 2004. Aquatic macrophytes potential for the simultaneous removal of heavy metals (Buenos Aires, Argentina). Chemospere, 57 (8): 997-1005.

Olivera, P., D. Backovic and S. Sladana, 2005. Dietary selenium supplementation of pigs and broilers as a way of producing selenium enriched meat. Acta Veterinaria (Beograd), 55 (5-6): 483-492.

Ortman, K. and B. Pehrson, 1999. Effects of selenate as feed supplement to dairy cows in comparison to selenite and selenium yeast. J. Anim. Sci., 77 (12): 3365-3370.

Pan, C., K. Huang, Y. Zhao, S. Qin, F. Chen and Q. Hu, 2007. Effect of selenium source and level in hen's diet on tissue selenium deposition and egg selenium concentration. J. Agric. Food Chem., 55 (3): 1027-1032.

Payne, R.L., T.K. Lavergne and L.L. Southern, 2005. Effect of inorganic versus organic selenium on hen production and egg selenium concentration. Poult. Sci., 84 (2): 232-237.

Pechova, A., L. Misurova, L. Pavlata and R. Dvorak, 2008. Monitoring of changes in selenium concentration in goat milk during short-term supplementation of various forms of selenium. Biol. Trace Elem. Res., 121 (2): 180-191.

Phalson, A.M., 1989. Toxicity of heavy metals (Zn, Cu, Cd, Pb) to vascular plants. Water Air Soil Pollut., 47 (3-4): 287-319.

Rao, P.N. and A.S. Reddy, 1984. Studies on the population biology of water lettuce: *Pistia stratiotes* L. Hydrobiologia, 119 (1): 15-19.

SAS, 1996. SAS/STAT® User's Guide (Release 6.03 ed.). SAS Inst. Inc. Cary, NC.

Satyakala, G. and K. Jamil, 1992. Chromium-induced biochemical changes in *Eichhornia crassipes* (Mart) *solms* and *Pistia stratiotes* L. Bull. Environ. Contam. Toxicol., 48 (6): 921-928.

Schrauzer, G.N., 2002. Selenium and human health: The relationship of selenium status to cancer and vital disease. Alltech's 18th Annual Symposium, pp: 263-269.

Skinner, K., N. Wright and E. Porter-Goff, 2007. Mercury uptake and accumulation by four species of aquatic plants. Environ. Pollut., 145 (1): 234-237.

Skrivan, M., J. Simane, G. Dlouha and J. Doucha, 2006. Effect of dietary sodium selenite, Se-enriched yeast and Se-enriched Chlorella on egg Se concentration, physical parameters on eggs and laying hens production. Czeck J. Anim. Sci., 51 (4): 163-167.

Sridhar, M.K.C., 1986. Trace element composition of *Pistia stratiotes* L. in a polluted lake in Nigeria. Hydrobiologia, 131 (3): 273-276.

Steel, R.G.D. and J.H. Torries, 1989. Principle and Procedure of Statistic a Biomaterial Approach. 2nd Edn. McGraw-Hill. New York, USA.

Sugihara, S., M. Kondo, Y. Chihara, M. Yuji, H. Hattori and M. Yoshida, 2004. Preparation of selenium-enriched sprouts and identification of their selenium species by high-performance liquid chromatography-inductively couple plasma mass spectrometry. Biosci. Biotechnol. Biochem., 68 (1): 193-199.

Suhajda, A., J. Hegoczki, B. Janzso, I. Pais and G. Vereczkey, 2000. Preparation of selenium yeast I. Preparation of selenium-enriched *Saccharomyces cerevisiae.* J. Trace Element Med. Biol., 14 (1): 43-47.

Tinggi, U., 2003. Essentiality and toxicity of selenium and its status in Australia: A review. Toxicol. Lett., 137 (1-2): 103-110.

Tsuneyoshi, T., J. Yoshida and T. Sasaoka, 2006. Hydroponic cultivation offers a practical means of producing selenium-enriched garlic. J. Nutr., 136 (3): 870S-872S.

Underwood, E.J. and N.F. Suttle, 1999. The Mineral Nutrition of Livestock. 3rd Edn. Biddles Ltd., Guildford and King's Lynn, Oxon, UK.

Xiong, Z.T., 1998. Lead uptake and effects on seed germination and plant growth in a Pb hyperaccumulator *Brassica pekinensis*. Rupr. Bull. Environ. Contam. Toxicol., 60 (3): 285-291.

Zhan, X.A., M. Wang, R.Q. Zhao, W.F. Li and Z.R. Xu, 2007. Effect of different selenium source on selenium distribution, loin quality and antioxidant status in finishing pigs. Anim. Feed Sci. Technol., 132 (3-4): 202-211.