

# THE SELENIUM UPTAKE IN TARTARY BUCKWHEAT SPROUTS GROWN FROM SEEDS SOAKED IN VARIOUS SELENIUM SOLUTIONS

Petra Štrekelj, Ivan Kreft, Vekoslava Stibilj

## ABSTRACT

The use of Tartary buckwheat (*Fagopyrum tataricum* Gaertn.) was declined for many years because of its bitter taste, but cultivation has raised with the awareness of high content of tannins and other polyphenols. Furthermore, tartary buckwheat is suitable for ecological growing, without the use of pesticides. Products made from buckwheat are floral honey, green buckwheat tea, buckwheat sprouts, and fresh green plant parts used as vegetable. Selenium (Se) is an essential trace element for mammals with a very narrow concentration range between insufficiency and toxicity. The availability and toxicity both depend on the chemical form of Se in food. The aim of this study was to determine the impact of various concentrations (5, 10 mg/l) of Se(IV) and Se(VI) in seed soaking solution on dry matter and its Se content in tartary buckwheat sprouts. Sensitive detection was achieved by HG-AFS with the chemicals and instrumental operating conditions according to Smrkoj and Stibilj (2004). Results shows that uptake of Se is higher in the form of sodium selenate than in the form of sodium selenite.

**Keywords:** glycinats copper, zinc, iron, cobalt and manganese, layer, blood, metabolism

## INTRODUCTION

Selenium is an essential trace element for humans and animals but for plants it has not been classified yet (Hartikainen, Xue, 1999; Hartikainen et al. 2000; Kuznetsov et al., 2003; Shanker, 2006). Selenium is a component of glutathion peroxidase enzyme (provides protection against oxidative stress) and many other Se-containing enzymes (thioredoxin reductase, iodothyronine deiodinase) (Driscoll et al., 2003).

Recommended daily allowance of selenium is for healthy adults 55 µgSe/day and pregnant women requires 60 µgSe/day. Selenium containing diet prevents against atherosclerosis, arthritis and cancer (Duffield-Lillico et al., 2003). Overdoses (3200-5000 µgSe/day) causes toxic reactions (selenosis) (Reid et al., 2004).

The major source of selenium is diet and the selenium level in the soil is reflected by the Se concentration in plant (Tingi, 2003). In Europe selenium level in soil is less than 1 mgSe/kg (Kadrabova et al., 1997), so the cultivation of plants enriched with selenium could be an effective way of producing Se-enriched foodstuffs, with benefits to health (Lyons et al., 2005; Germ et al. 2005; Germ et al. 2007ab). In foods, the bioavailability and toxicity of Se depend on its chemical form (Mazej et al., 2006).

Tartary buckwheat (*Fagopyrum tataricum* Gaertn.) in an important alternative crop because of high content of proteins, fiber, vitamins B1, B2, B6 (Bonafaccia et al. 2003 a,b; Smrkoj et al., 2006; Vogrinčič et al., 2009), and has more antioxidants (rutin) in comparison to common buckwheat (Fabjan et al., 2003).

Tartary buckwheat is used as sprouts, leafy vegetable, tea or flour from milled grain. It is free of content of gluten and thus safe for celiac disease patients (Bonafaccia et al., 2003 a,b).

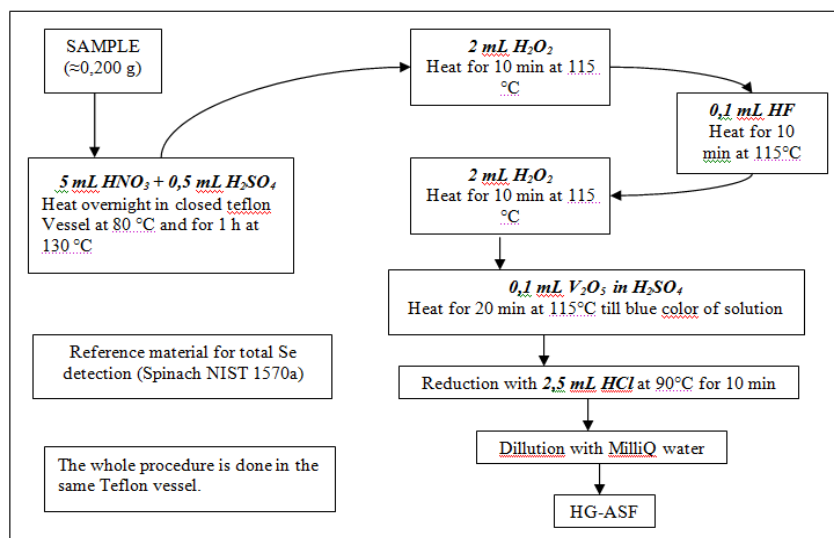
The aim of this study was to determine the impact of various concentrations (5, 10 mgSe/L) of Se(IV) and Se(VI) in seed soaking solution on dry matter and Se content in tartary buckwheat sprouts.

## MATERIAL AND METHODOLOGY

Tartary buckwheat seeds obtained from Luxemburg were soaked (4 hours) in solution of either sodium selenate (Na<sub>2</sub>SeO<sub>4</sub>) or sodium selenite (Na<sub>2</sub>SeO<sub>3</sub>), dissolved in MilliQ water in concentrations 5 and 10 mgSe/L. Seeds were sown in bowls on wet towel paper and exposed to daylight without UV radiation (in greenhouse). Sampling was done when the sprouts developed two extended cotyledon leaves. Samples were lyophilized and milled.

### Selenium determination

Each sample was analysed at least in triplicate. Briefly, 0.5 mL concentrated H<sub>2</sub>SO<sub>4</sub> and 1.5 mL concentrated HNO<sub>3</sub> were added to 0.2 g of sample in a Teflon tube, which was closed and heated in an aluminium block at 80°C overnight, and than for 1 h at 130 °C. After cooling, 2 mL of H<sub>2</sub>O<sub>2</sub> was added and the tubes were heated for 10 min at 115°C. Because samples also contained fibres 0.1 mL 40% HF was added and heated at 115°C for 10 min. After that the step with H<sub>2</sub>O<sub>2</sub> was repeated. After the solution had cooled to room temperature, 0.1 mL V<sub>2</sub>O<sub>5</sub> in H<sub>2</sub>SO<sub>4</sub> was added and the tube heated at 115°C for 20 min (until the solution became blue in colour). The reduction of Se(VI) to Se(IV) was obtained by the addition of concentrated HCl and heating at 90°C for 10 min (Fig 1). Samples were diluted with Milli Q



water before determining Se content. Working standard solutions were prepared daily in the range from 0.1-10 ng/g. The standards were prepared in the same media as samples. To check the accuracy and precision of the method a standard reference material representing a similar matrix was analysed simultaneously. Se content was detected with HG-AFS (Fig. 2).

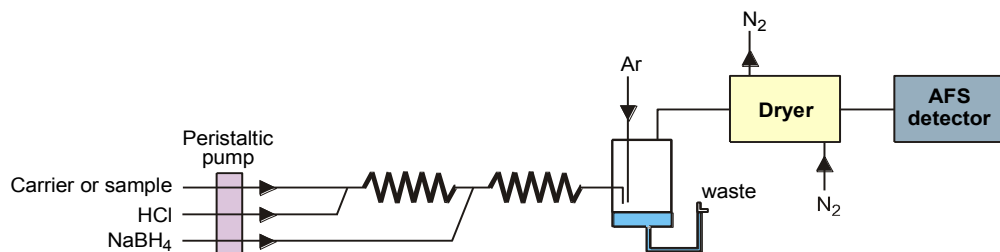


Fig. 2: Selenium detection with HG-AFS

**Selenium detection**

A continuous flow rate hydride generation atomic fluorescence spectrometer equipped with selenium cathode lamp, was employed for HG-AFS. The gaseous hydrogen selenide was formed after mixing and reaction of the solution (or carrier (0.05 mol/L HCl, 1 mL/min)) with NaBH<sub>4</sub> (1.2% (mV-1) in 0.1 mol/L NaOH at a flow rate of 3 mL/min and HCl (2 mol/ L 1.8 mL/min) in a polyetheretherketone (PEEK) cross-connector. The mixture of H<sub>2</sub> and H<sub>2</sub>Se was separated by a gas-liquid separator, then transferred with a flow of argon (0.260L/min) through the dryer to the AFS detector. Ar is carrier gas and it is inert to H<sub>2</sub>Se, the dryer gas was nitrogen (3 L/min). Atomic fluorescence signals were registered and measured on the basis of peak height, in nm. Flow rates were adjusted at a peristaltic pump by using tubing with different internal diameters (Smrkolj, Stibilj; 2004).

Table 1: Selenium detection with HG-AFS

Parameter	Value
carrier flow rate	1 mL/min
argon flow rate	0,260 L/min
nitrogen flow rate	3L/min
conc. of NaBH <sub>4</sub>	1,2% w/v
conc. of HCl for HG	2 mol/L
conc. of HCl in carrier	0,05 mol/L

**RESULTS AND DISCUSION**

During the plant growth no toxic signs were noticed. Sprouts dry mass raised with the addition of selenium solution (Table 1). Se (VI) has higher impact on the sprouts dry mass than Se (IV).

Se content was dependent on the form and concentration of soaking solution. The higher amount of selenium was in the sprouts which seeds were soaked in sodium selenate solution, here the content of Se was 2-3x higher that in sprouts from seeds soaked in selenite solution. The dependence between concentration of soaking solution and Se content obtained is almost linear.

The percentage of Se uptake in buckwheat sprouts was calculated from the ratio between the measured mass of Se in the sprout (Se concentration in sprout multiplied by the average sprout dry mass) and the theoretical one in the seed. Se uptake increase almost linearly with concentration of soaking solution and it is higher in selenate soaking solution.

Table 2: Tartary buckwheat sprouts

Soaking solution	Concentration (mgSe/L)	Average DM of sprout(g)	Se content (µg/g)	Se uptake (%)
Water	0	0.00852±0.073	0.053±0.054	
Se(VI)	5	0.0091±0.047	4.835±0.897	54
	10	0.0085±0.051	10.839±2.434	56
Water	0	0.00797±0.053	0.062±0.036	
Se(IV)	5	0.0088±0.068	1.225±0.214	13
	10	0.0086±0.043	3.415±0.488	18

Table 3: Tartary buckwheat sprouts

Soaking solution	Concentration (mgSe/L)	Average DM of sprout(g)	Se content (µg/g)	Se uptake (%)
Water	0	0.097±0.006	0.050±0.001	
Se(VI)	5	0.0107±0.005	4.03±0.11	58
	10	0.0093±0.009	9.67±0.11	62
Water	0	0.114±0.004	0.071±0.002	
Se(IV)	5	0.0081±0.003	2.39±0.22	27
	10	0.0066±0.002	2.59±0.11	12

**CONCLUSION**

The Se (VI) and Se (IV) soaking solution has a positive impact on sprouts growth, and Se(VI) has the higher impact on the sprouts dry mass than Se (IV). Sprouts dry mass raised by increasing concentration of Se in soaking solution. Se content was dependent on the form and concentrations of Se soaking solution. Sprouts were uptaking the higher amount of Se in the form of sodium selenate than in the form of sodium selenite.

REFERENCES

- BONAFACCIA, G., GAMBELLI, L., FABJAN, N., KREFT, I. 2003a. Trace elements in flour and bran from common and tartary buckwheat. In *Food Chemistry*, vol. 83, p. 1-5.
- BONAFACCIA, G., MAROCCHINI, M., KREFT, I. 2003b. Composition and technological properties of the flour and bran from common and tartary buckwheat. In *Food Chemistry*, vol. 80, p. 9-15.
- CUDERMAN, P., OŽBOLT, L., KREFT, I., STIBILJ, V. 2010. Extraction of Se species in buckwheat sprouts grown from seeds soaked in various Se solutions. In *Food Chemistry*, vol. 123, p. 941-948.
- DRISCOLL, D. M., COPELAND, P. R. 2003. Mechanism and regulation of selenoprotein synthesis. In *Ann. Rev. Nutr.*, vol. 23, p. 17-40.
- DUFFIELD – LILLICO, A. J., DALKIN, B. L., REID, M. E., TURNBULL, B. W., SLATE, E. H., JACOBS, E. T., MARSHALL, J. R., CLARK, L. C. 2003. Selenium supplementation, baseline plasma selenium status and incidence of prostate cancer: an analysis of the complete treatment period of the Nutritional Prevention of Cancer Trial. *BJU*, vol. 91, p. 608-612.
- FABJAN, N., RODE, J., KOŠIR, I. J., WANG, Z., ZHANG, Z., KREFT, I. 2003. Tartary buckwheat (*Fagopyrum tataricum* Gaertn.) as a source of dietary rutin and quercitrin. In *Journal of Agricultural and Food Chemistry*, vol. 51, p. 6452-6455.
- GERM, M., KREFT, I., OSVALD, J. 2005. Influence of UV-B exclusion and selenium treatment on photochemical efficiency of photosystem II, yield and respiratory potential in pumpkins (*Cucurbita pepo* L.). In *Plant Physiology and Biochemistry*, vol. 43, p. 445-448.
- GERM, M., STIBILJ, V., KREFT, I. 2007a. Metabolic importance of selenium for plants. In *The European Journal of Plant Science and Biotechnology*, vol. 1, p. 91-97.
- GERM, M., STIBILJ, V., OSVALD J., KREFT, I. 2007b. Effect of selenium foliar application on Chicory (*Cichorium intybus* L.). In *Journal of Agricultural and Food Chemistry*, vol. 55, p. 795-798.
- HARTIKAINEN H., XUE T., 1999. The promotive effect of selenium on plant growth as triggered by ultraviolet irradiation. In *J. Environ. Qual.*, vol. 28, p. 1272-1275.
- HARTIKAINEN, H., XUE, T., PIIRONEN, V. 2000. Selenium as an antioxidant and pro-oxidant in ryegrass. In *Plant and Soil*, vol. 225, p. 193-200.
- KADRABOVA, J., MADARIC, A., GINTER, E. 1997. The selenium content of selected food from Slovak Republic. In *Food Chemistry*, vol. 58, p. 29-32.
- KUZNETSOV, V. V., KHOLODOVA, V. P., KUZNETSOV, V. I. V., YAGODIN, B. A. 2003. Selenium regulates the water status of plants exposed to drought. In *Doklady Biological Sciences*, vol. 390, p. 266-268.
- LYONS, G., ORTIZ-MONASTERIO, I., STANGOULIS, J., GRAHAM, R. 2005. Selenium concentration in wheat grain: Is there sufficient genotypic variation to use in breeding? In *Plant and Soil*, vol. 269, p. 369-380.
- MAZEJ, D., FALNOGA, I., VEBER, M., STIBILJ, V. 2006. Determination of selenium species in plant leaves by HPLC-UV-HG-AFS. In *Talanta*, vol. 68, p. 558-568.
- REID, M. E., STRATTON, M. S., LILLICO, A. J., FAKIH, M., NATARAJAN, R., CLARK, L. C., MARSHALL, J. R. 2004. A report of high-dose selenium supplementation: response and toxicities. In *Journal of Trace Elements in Medicine and Biology*, vol. 18, p. 69-74.
- SHANKER, A. K. 2006. Countering UV-B stress in plants: does selenium have a role? In *Plant and Soil*, vol. 282, p. 21-26.
- TINGGI, U. 2003. Essentiality and toxicity of selenium and its status in Australia: a review. In *Toxicology Letters*, vol. 137, p. 103-110.
- SMRKOLJ, P., STIBILJ, V. 2004. Determination of selenium in vegetables by hydride generation atomic fluorescence spectrometry. In *Analytica Chimica Acta*, vol. 512, p. 11-17.
- SMRKOLJ, P., STIBILJ, V., KREFT, I., GERM, M. 2006. Selenium species in buckwheat cultivated with foliar addition of Se(VI) and various levels of UV-B radiation. In *Food Chemistry*, vol. 96, p. 675-681.
- VOGRINČIČ, M., CUDERMAN, P., KREFT, I., STIBILJ, V. 2009. Selenium and its species distribution in above-ground plant parts of selenium enriched buckwheat (*Fagopyrum esculentum* Moench.) In *Analytical Sciences*, vol. 25, p. 1357-1363.

**Acknowledgments:**

The study was financed by the following programs and projects of the Slovenian Research Agency: P4-0085 and J4-3618, Tartary buckwheat as a novel source of functional foods, and Fellowship for “Young Researchers” to Petra Štrekelj.

**Contact address:**

Petra Štrekelj, University of Ljubljana, Biotechnical Faculty, Jamnikarjeva 101, SI-1000 Ljubljana, Slovenia, e-mail: petra.strekelj@gmail.com

Ivan Kreft, University of Ljubljana, Biotechnical Faculty, Jamnikarjeva 101, SI-1000 Ljubljana, Slovenia, e-mail: ivan.kreft@guest.arnes.si

Vekoslava Stibilj, Inštitut “Jožef Stefan”, Jamova 39, SI-1000 Ljubljana, Slovenia, e-mail: vekoslava.stibilj@ijs.si