

Biofortification of food chain with selenium

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Abstract

Selenium (Se) is an essential micronutrient for humans and animals which is circulated to food chain via crop plants. Agronomic biofortification with Se is used in areas where the soil Se content is low and Se deficiency causes health problems. *Brassica* species are efficient in accumulation of soil Se and therefore an attractive choice of species for biofortification. Se uptake and translocation was studied in *B. rapa* and *B. napus* in field experiments applied with 0, 6 or 20 mg Se ha⁻¹ as Na₂SeO₄ and foliar sprays of 30 mg Se ha⁻¹ as Na₂SeO₄ or Na₂SeO₃. In addition, a HPLC-ICP-MS based method to identify and quantify organic Se-compounds in seeds and meals was developed. High concentration of Se accumulated in seeds (1.89 µg g⁻¹) and after oil extraction Se remained mainly in the meal fraction (1.34 µg g⁻¹). Foliar spray of Na₂SeO₄ was most efficient in increasing the meal Se concentration. Selenium specification studies revealed that 68-82 % of Se was in a form of valuable selenomethionine (SeMet). Other Se-compounds were not detected. At rosette stage, *Brassica* plants had taken up 30-40% of applied Se but the translocation to seeds was low. The results show that agronomic biofortification with Se can improve the nutritive quality of *Brassica* meal due to high accumulation of SeMet and its stability during processing. The strategy to improve the efficiency of agronomic biofortification with Se in Brassica is discussed.

Introduction

Selenium (Se) is recognized as an essential microelement for human and animals due to its antioxidative properties and role in hormone balance. In plants, evidence on the beneficial effects of Se is increasing (Hartikainen 2005). Both too low and high Se concentrations in the food chain can cause health problems and eventually be lethal to humans and animals. Selenium is circulated to food chain mainly via crops and therefore the Se status of the food chain is dependent on Se level of the soil. In Finland, where soils are deficient in Se, the recommended daily intake of Se has been

secured by agronomic biofortification e.g. application of Se in fertilizers (Hartikainen 2005). Genetic biofortification e.g. breeding for improved Se concentration have not been used due to lack of genetic variation in Se density in modern wheat (Lyons et al. 2005) and low acceptance of genetically modified crops. Also, the distribution of Se in earth crust is uneven, and areas of Se deficiency and toxicity can locate close to each other.

The inorganic Se compounds applied in fertilizers are taken up by plants and assimilated into valuable organic Se-compounds (Pilon-Smits et al. 2009), which are safer and their retention in tissue is better (Rayman 2004). Some plant species such as *Astragalus* sp. and *Brassica* sp. can also assimilate bioactive Se-compounds such as a precursor of a chemotherapeutic compound, selenomethyl Se-cysteine (SeMeSeCys). The reported additional health benefits of Se, such as improved immune system and reduced cancer risk, require higher than recommended daily intakes and therefore feed additives such as Se enriched yeast (SY), containing approximately 80 % of selenomethionine (SeMet), have been developed (Rayman 2004). The ability to accumulate and assimilate Se varies between plant species, dividing them into hyperaccumulators (e.g. *Astragalus*), Se accumulators (e.g. *Brassica juncea*) and non-accumulators (e.g. wheat) (Pilon-Smits et al. 2009). According to current knowledge the same transporters that are active in sulphur (S) uptake and metabolism are also responsible for Se metabolism (Pilon-Smits et al. 2009). *Brassica* species, which are efficient in both S and Se accumulation and which are used as protein source in ruminant diets, are therefore attractive choice of crops for the biofortification of food chain with Se.

Materials and methods

Field experiments were carried out at Viikki Experimental Farm, University of Helsinki, Finland (EXP1). *B. rapa* (Hilight) and *B. napus* (4021B) were sown in separate experiments in a completely randomized block design with four replications. All plots were fertilized at sowing with 100 kg N ha⁻¹ three application levels of Se 0, 5.6 and 30 g Se ha⁻¹ as Na₂SeO₄. The composition of used fertilizers were; N27-P0-K0 + Mg 4.0 % + Se 0, N27-P0-K0 + Mg 1 % + B 0,02 % + Se 0.0015% and N26-P0-K1 + S 3 % + Mg 1 % + B 0,02 % + Se 0,005%. A foliar spray of Na₂SeO₄ or Na₂SeO₃ of 0

and 30 g Se ha⁻¹ was applied a month after the sowing at rosette stage (BBCH 30-33). Petroleum ether was used for oil extraction and the residual meal and seeds were analyzed for total Se content (Kumpulainen et al. 1983). Separation, identification and quantification of Se species was carried out by using HPLC-ICP-MS as described in (Seppänen ym., manuscript). The uptake and translocation of Se was quantified by measuring the Se concentration of *Brassica* foliage before and after foliar Se application and that of seeds, pods, residual plant material and roots during harvest.

Results and discussion

Both *Brassica* species accumulated high amounts of Se in the protein fraction (meal) of the seed and up to 85 % of Se was in the form of SeMet (Table 1). SeMet being the main Se species both in meal and seeds implies that processing of *Brassica* seed to defatted meal does not alter SeMet originally present in seeds. The percentage of SeMet of total Se was at same ratio as in the selenised yeast used in feed additives (see e.g. Rayman 2004).

Table 1. Concentration of total Se ($\mu\text{g/g}$), SeMet ($\mu\text{g/g}$), inorganic Se(IV) ($\mu\text{g/g}$) and the ratio between SeMet and total Se (%) the *Brassica* meal of plants supplemented with Na₂SeO₄ in soil Se0, Se6 and Se20 (0, 6 or 20 mg Se ha⁻¹) and leaves as Na₂SeO₄ (1) or Na₂SeO₃ (2) (20 mg Se ha⁻¹) in location EXP1.

	<i>B.rapa</i>					<i>B.napus</i>				
	Se	SeMet	Se(VI)	%	R	Se	SeMet	Se(VI)	%	R
Se0	0.05	0.03	0.00	73	80.2	0.06	0.04	0.00	78	83.6
Se0+1	1.14	0.86	0.06	76	80.5	1.24	0.79	0.03	64	66.3
Se0+2	0.33	0.24		72	72.5	0.25	0.19		77	77.0
Se6	0.27	0.20		74	74.2	0.24	0.20		82	81.3
Se6+1	1.45	1.00		68	68.4	1.34	0.95		71	70.7
Se6+2	0.57	0.39	0.06	68	78.0	0.43	0.30	0.03	70	76.7
Se20	0.78	0.57	0.04	73	77.7	0.82	0.58	0.03	71	75.1
Se20+1	1.96	1.37		70	69.7	1.45	1.20		82	61.2
Se20+2	0.93	0.75		80	80.3	1.04	0.78		75	75.0

The SeMet content in meal increased to 0.86 $\mu\text{g g}^{-1}$ when Na₂SeO₄ was applied as foliar spray and the soil application of 6 or 20 mg Se ha⁻¹ elevated the level further to 1.00 and 1.37 $\mu\text{g g}^{-1}$, respectively. Without Se application, only traces of SeMet could be detected in the meal. The Se-enriched *Brassica* meal can be used as protein source for up to 50 % in feed concentrates and it can significantly add up the amount of

SeMet in ruminant diet. Significant proportion (30-40%) of applied Se was found in foliage at rosette stage (Table 2) and the Se concentration of foliage, meal and residue correlated with the Se application level. The translocation to seeds was below 10 %, lower than reported for field grown wheat. High capacity to volatilize methylated Se-compounds is characteristic for *Brassica* sp. (Pilon-Smits et al. 2009) and may explain the loss of Se.

Table 2. The Se ($\mu\text{g Se g}^{-1}$) of *Brassica rapa* and *B. napus* foliage, meal and residual plant material containing pods and straw. Foliage samples were harvested at two stages of development before (1st) and after (2nd) Se foliar sprays. See Table 1.

	<i>B.rapa</i>				<i>B.napus</i>			
	1 st	2 nd	Meal	Residue	1 st	2 nd	Meal	Residue
Se0	0.04	0.03	0.05 ^f	0.03	0.09 ^d	0.05 ^d	0.05 ^e	0.02 ^c
Se0+1	0.05	5.47	1.00 ^{bc}	0.48	0.15 ^{bc}	6.52 ^a	0.94 ^b	0.40 ^{ab}
Se0+2	0.05	1.39	0.23 ^{ef}	0.15	0.10 ^{bc}	0.70 ^{cd}	0.18 ^e	0.08 ^{bc}
Se6	0.27	0.80	0.23 ^{ef}	0.14	0.17 ^{abc}	0.91 ^{cd}	0.24 ^{de}	0.09 ^{bc}
Se6+1	0.35	7.22	1.15 ^b	0.63	0.15 ^{abc}	5.83 ^{ab}	1.07 ^{ab}	0.53 ^a
Se6+2	0.24	1.64	0.43 ^{def}	0.23	0.10 ^{abc}	1.67 ^{cd}	0.38 ^{cd}	0.14 ^{bc}
Se20	1.19	3.30	0.60 ^{cde}	0.28	0.49 ^a	3.37 ^{bc}	0.68 ^{bc}	0.37 ^{ab}
Se20+1	0.98	8.60	1.84 ^a	0.86	0.43 ^{ab}	8.70 ^a	1.48 ^a	0.68 ^a
Se20+2	0.82	4.68	0.79 ^{bcd}	0.50	0.37 ^{abc}	3.38 ^{bc}	0.75 ^{bc}	0.39 ^{ab}

Conclusion

Our results show that the agronomic biofortification with Se can improve the nutritional quality of *Brassica* meal and the capacity of *Brassica* species to accumulate Se gives an attractive option for increasing the SeMet concentration of animal diets.

References

- Hartikainen H. 2005. Biogeochemistry of selenium and its impact on food chain quality and human health. *J. Trace Elem. Med. Biol.* 18:309-318.
- Lyons G, Ortiz-Monasterio I, Stangaoulis J, Graham R 2005. Selenium concentration in wheat grain: is there sufficient genotypic variation to use in breeding. *Plant Soil* 269:282-292.
- Pilon-Smits EAH, Quinn CF, Tapken W, Malagoli M, Schiavon M 2009. Physiological functions of beneficial elements. *Curr. Opin. Plant Biol.* 12:267-274.
- Rayman M. 2004. The use of high-Se yeast to raise Selenium status – how does it measure up? *British J. of Nurt.* 92:557-573.