

SELENIUM SUPPLEMENT AND CYS/MET INDEX IN TARM 92 BARLEY CULTIVAR

Semih Yılmaz^{1*}, Ali İrfan İlbaş², Aysun Çetin³, Aydın Uzun⁴, Hasan Pınar⁴, Svetlana Garaeva⁵

¹ Erciyes University, Faculty of Agriculture, Department of Agricultural Biotechnology, Kayseri, Turkey,

² Erciyes University, Faculty of Agriculture, Department of Field Crops, Kayseri, Turkey,

³ Erciyes University, Faculty of Medicine, Department of Medical Biochemistry, Kayseri, Turkey,

⁴ Erciyes University, Faculty of Agriculture, Department of Horticulture, Kayseri, Turkey,

⁵ Academy of Sciences of Moldova, Chisinau, Moldova

Abstract

Selenium (Se) as essential micronutrient found in foods in the form of organic selenomethionine (SeMet) and selenocysteine (SeCys) in adequate amounts. However, selenite (SeO_3^{2-}) or selenate (SeO_4^{2-}) can be found in inorganic form in very low amounts. Higher plants indicate considerable variations in terms of Se requirement with various effects on plant metabolism and uptake of some nutrients. Barley (*Hordeum vulgare* L.) is a significant dietary source for this element for humans and animals. In present study, Cys/Met index of Tarm 92 registered barley cultivar subjected to increasing sodium selenate (Na_2SeO_4) doses (6.25, 12.50, 18.75, 25.00 g ha⁻¹) was investigated. Amino acids were analyzed according to Ion Exchange Liquid Chromatography method by Amino Acid Analyzer AAA 339 M. The experimental soil was analyzed for texture, organic matter, pH, available Phosphorus (P), Nitrogen (N), and Potassium (K). The average Sulfur (S) level in Tarm 92 cultivar was 0.0514 mg 100mg⁻¹. Results revealed significant effects of Se-treatments on grain Cys/Met index at 12.50 g ha⁻¹. Average Cys/Met index remarkably increased to highest level at 12.50 g ha⁻¹. However, at 25 g ha⁻¹, the level (6.98 mg 100mg⁻¹) was closer to that in control (5.10 mg 100mg⁻¹) group. It was concluded in present study that 12.5 g ha⁻¹ selenium treatment was sufficient to increase grain Cys/Met index of Tarm 92 barley cultivar.

Keywords: Barley, Cys/Met index, Selenium, Sulfur, Tarm 92.

1. INTRODUCTION

Main dietary source of selenium (Se) for both humans and animals are plants. Therefore, knowledge of Se compounds in plants are of crucial importance (Dumont et al., 2006). In this manner Barley is one of the most important grains used as animal feed. Components and amino acid compositions of cereal grains are important quality criteria for nutritional value to animals (Assveen, 2009). For this reason, protein content of barley, used as animal feed, has to be high enough.

Selenium (Se) is physically and chemically similar to Sulfur (S) and can replace S in amino acids (Levander and Burk, 1994; Sors et al., 2005). S is very important for proteins as part of the characteristic component of methionine (Met) and cysteine (Cys) in organisms (Mortensen et al., 1992). Therefore, it is known that S leads to an increase in Cys/Met index in barley (Mortensen et al., 1992). In addition, Se enriched barley may provide additional compounds for livestock feeding (İlbas et al., 2012). In organisms, Se is specifically believed to shows its effect by incorporating into proteins as the amino acid selenocysteine (SeCys) (Behne and Kyriakopoulos, 2001). It can also

replace S in methionine (Met), forming selenomethionine (SeMet) and be included as part of proteins (Behne and Kyriakopoulos, 2001).

Met is prerequisite for initiating the protein synthesis via addition of aa's one after another in the chain, and Cys is responsible for the formation of disulfate bridges between the polypeptides to make them functional (Mortensen et al., 1992). The physical and chemical similarities between sulfur and selenium explains the relationship between these two elements in terms of their metabolic usage in plants (Sors et al., 2005). Plants predominantly uptake S and Se in the form of sulfate, selenate and selenite ions. Biological importance of Se as nutrient is well known in terms of development of many living species (Fishbein, 1991; Cutter and Bruland, 1984). On the other hand, it can also exhibit toxic effects on organisms (Merian and Clarkson, 1991). The non-specific integration of the seleno-amino acids SeCys and SeMet into proteins is believed to be the major contributor of Se toxicity in plants (Brown and Shrift, 1981, 1982).

In the present study, considering the difficulties of some other approaches to improve the nutritional quality of grains, the effect of Se fertilization was tested on grain Cys/Met index and thousand grain weights of Tarm 92 barley cultivar. This study aimed to indicate the association between Cys/Met index and yield to take an initial look at the effect of Sodium selenate (Na_2SeO_4) concentrations on Tarm 92 cultivar.

2. MATERIALS AND METHODS

Registered Tarm-92 barley cultivar was used as test material. The study was designed in three replications for every concentration of Na_2SeO_4 . In each replication 5 parcels (2 m^2) were prepared and Na_2SeO_4 concentrations were applied (6.25, 12.50, 18.75, 25 g ha^{-1}) simultaneously with sowing the seeds. After 4 months of maturation period, plants were uprooted and packed for Cys/Met and mineral analysis.

Analytical determination

Sample preparation was made by the recommendations from standard operation procedures of EPA, PerkinElmer Inc., USGS (Perkin Elmer, 2000). Se concentrations in all samples were analyzed by hydride generation Atomic Absorption Spectrophotometry (FIAS-400, Perkin Elmer AAnalyst800). The methods detection limit was 0.003 mg kg^{-1} . S contents of grain samples of the barley cultivars were examined using the method of Butters and Chenery (1959).

Cys/Met index analysis

Cys and Met were determined by ion exchange liquid chromatography using AAA 339 M amino acid analyzer (OSTION LG ANB column, 8 mm diameter, 35 mm length) as in our previous study (Yilmaz et al., 2017). Colorimetric determination of colored complexes formed by reaction with ninhydrin was performed at 570 nm. Analysis of quantitative measurements was performed by automated determination of peak areas for identified acids. The Cys-Met index is a simple ratio of cysteine to methionine and is calculated using the concentrations of amino acid Cys divided by Met value from the same sample.

Statistical Analysis

A two-way analysis of variance (Two-way ANOVA) was performed to investigate the effects of sodium selenate doses on Cys/Met index of Tarm 92 cultivar using SPSS for windows (SPSS, Inc., Chicago, IL, USA). All values are expressed as mean and standard deviation statistics. $P < 0.05$ probability level was considered as statistically significant.

3. RESULTS AND DISCUSSION

Experimental soil (pH, 7.83) contained 0.8 mg kg^{-1} Se, 99 mg g^{-1} K, 0.21% N, and 5 mg g^{-1} P, and 2.5% organic matter. According to the range determined by FAO, the content of the soil organic matter was sufficient, but Se content of experimental soil (0.8 mg kg^{-1}) was higher than that of World's mean (0.4 mg kg^{-1}) (Sillanpaa, 1990; Ilbas et al., 2012). Amount of grain Se at doses 0.00, 6.25, 12.50, 18.75 and 25.00 g ha^{-1} was calculated as 0.096, 0.158, 0.132, 0.126 and 0.079 mg kg^{-1} , respectively. The values at mid doses were significantly higher compared to control. At the same doses the grain S content was 0.0394, 0.0589, 0.0462, 0.0627 and $0.0496 \text{ mg } 100\text{mg}^{-1}$, respectively (Figure 1).

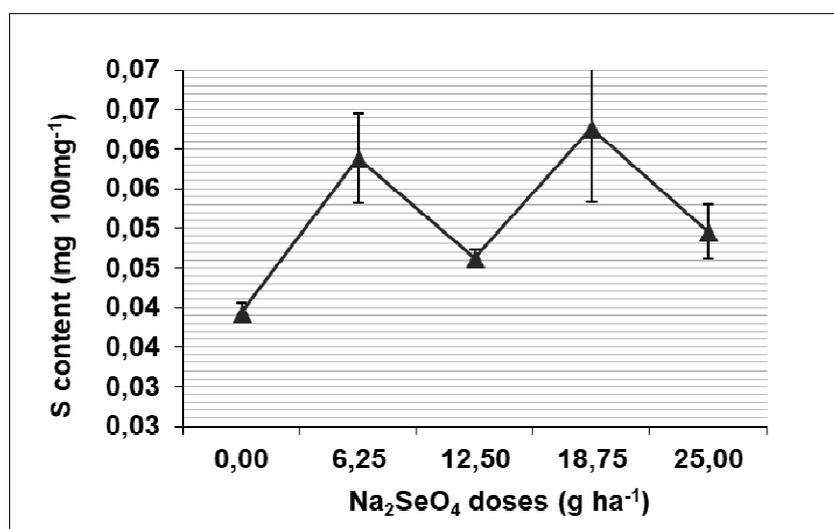


Figure 1. Sulfur content in grains

Contrary to Se content, S was at the lowest level at mid dose (12.5 g ha^{-1}). However, Cys/Met index at 12.50 g ha^{-1} indicated a remarkable increase and reached to highest level ($P < 0.05$). Subsequently, the level at doses 18.75 and 25 g ha^{-1} was observed as 5.90 and $6.98 \text{ mg } 100\text{mg}^{-1}$ with insignificant decrease at 12.50 g ha^{-1} (Figure 2).

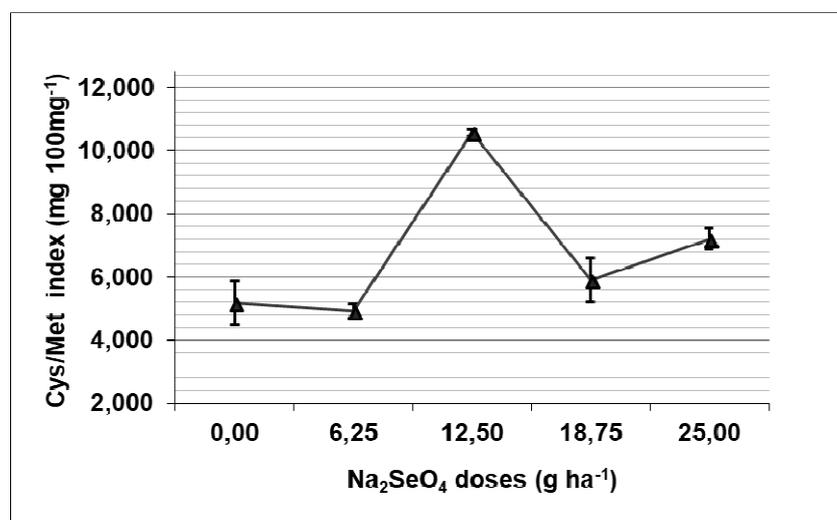


Figure 2. Cys/Met index in grains

For the thousand grain weight, significant difference was not observed after application of Se doses, but the response trend of the cultivar was similar as in Cys/Met index (Figure 3). Contrarily, response trend for the S at all doses was just the opposite of thousand grain weight and Cys/Met index.

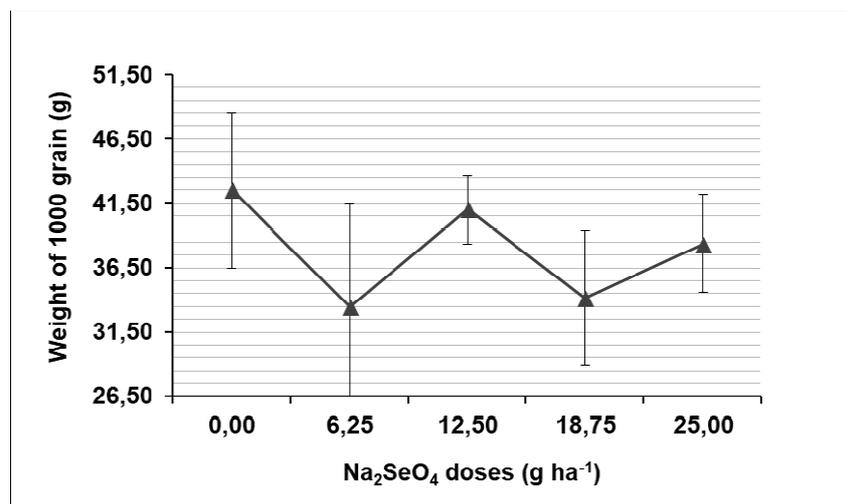


Figure 3. Thousand grain weight

Simojoki, et al., (2003) reported that while small Se additions increased Se contents in lettuce shoots and enhanced plant growth, Se contents above 20 mg kg⁻¹ caused a drastic drop in yield. Similar result was reported by Tamas et al., (2010) stating that depending on the increasing doses, a numeric decrease was observed in yield. In plant tissues, selenium enters into metabolism via sulfate assimilation pathway and cause changes in methionine and cysteine in proteins, often with a negative effect (Tamas et al., 2010). The reason for this situation might be that the major mechanism of Se toxicity in plants was the non-specific incorporation of selenocysteine and selenomethionine into proteins in place of Cys and Met, resulting in the alteration of protein structure (Tamas et al., 2010). On the other hand, Singh et al., (1980) reported the promoting effect of Se on plant growth, stating that 0.5 mg kg⁻¹ Se as Na₂SeO₃ stimulated growth and increased the dry weight of Indian mustard (*Brassica juncea* L.). Also, it was indicated that Se fertilization at low concentrations enhanced growth of both mono and dicotyledonous plants (Hasanuzzaman and Fujita, 2011). In accordance with this, in the present study, it was indicated that at mid dose (12.50 g ha⁻¹) Cys/Met index reached the highest level, but conversely S accumulation decreased to the lowest level. However, thousand grain weight was nearly at the same level as in control, while lower at other doses suggesting that yield and the Cys/Met index varies with a similar response.

4. CONCLUSION

It was concluded in present study that 12.5 g ha⁻¹ selenium treatment was sufficient to increase grain Cys/Met index to maximal level, but a noteworthy relationship was not observed between yield and Cys/Met index in Tarm 92 barley cultivar.

5. REFERENCES

Assveen, M. (2009). Amino acid composition of spring barley cultivars used in Norway. *Acta Agriculturae Scandinavica, Section B - Plant Soil Science*, 59(5), 395–401.

- Behne, D., Kyriakopoulos, A. (2001). Mammalian selenium-containing proteins. *Annual Review of Nutrition*, 21(1), 453–473.
- Brown, T.A., Shrift, A. (1981). Exclusion of selenium from proteins of selenium-tolerant astragalus species. *Plant Physiology*, 67(5), 1051–1053.
- Brown, T.A., Shrift, A. (1982). Selenium: toxicity and tolerance in higher plants. *Biological Reviews*, 57(1), 59–84.
- Butters, B., Chenery, E.M. (1959). A rapid method for the determination of total sulphur in soils and plants. *The Analyst*, 84(997), 239.
- Cutter, G.A., Bruland, K.W. (1984). The marine biogeochemistry of selenium: A re-evaluation 1. *Limnology and Oceanography*, 29(6), 1179–1192.
- Dumont, E., Vanhaecke, F., Cornelis, R. (2006). Selenium speciation from food source to metabolites: a critical review. *Analytical and Bioanalytical Chemistry*, 385(7), 1304–1323.
- Fishbein, L. (1991). Selenium. In E. Merian, eds, *Metals and their compounds in the environment, occurrence, analysis and biological relevance* (Ver. 2, pp. 1153-1190). Weinheim: VCH, Weinheim.
- Hasanuzzaman, M., Fujita, M. (2011). Selenium pretreatment upregulates the antioxidant defense and methylglyoxal detoxification system and confers enhanced tolerance to drought stress in rapeseed seedlings. *Biological Trace Element Research*, 143(3), 1758–1776.
- Ilbas, A.I., Yilmaz, S., Akbulut, M., Bogdevich, O. (2012). Uptake and distribution of selenium, nitrogen and sulfur in three barley cultivars subjected to selenium applications. *Journal of Plant Nutrition*, 35(3), 442–452.
- Levander, O.A., Burk R.F. (1994). Selenium. In M.E. Shils, J.A. Olson, M. Shike, eds, *Modern nutrition in health and disease*. Philadelphia: Lea and Febiger, Philadelphia.
- Singh, M., Singh, N., Bhandari, D.K. (1980). Interaction of selenium and sulfur on the growth and chemical composition of raya. *Soil Science*, 129(4), 238–244.
- Merian, E., Clarkson, T.W. (1991). *Metals and their compounds in the environment*: occurrence, analysis, and biological relevance. (Ver. 2, pp. 1438). Weinheim: VCH, Weinheim.
- Mortensen, J., Eriksen, J., Nielsen, J.D. (1992). Sulfur deficiency and amino acid composition in seeds and grass. *Phyton*, 3(3), 85-90.
- Perkin Elmer I., (2000). *Analytical Methods: Atomic Absorption Spectroscopy; Flow Injection Mercury/Hydride Analyses; THGA Graphite Furnace Recommended Analytical Conditions*. Waltham, MA, Perkin Elmer.
- Simojoki, A., Xue, T., Lukkari, K., Pennanen, A., Hartikainen, H. (2003). Allocation of added selenium in lettuce and its impact on roots. *Agricultural and Food Science in Finland*, 12, 155–164.
- Sors, T.G., Ellis, D.R., Salt, D.E. (2005). Selenium uptake, translocation, assimilation and metabolic fate in plants. *Photosynthesis Research*, 86(3), 373–389.
- Tamas, M., Mandoki, Z., Csapo, J. (2010). The role of selenium content of wheat in the human nutrition. A literature review. *Acta Univ. Sapientiae, Alimentaria*, 3, 5–34.
- Yilmaz, S., Ilbaş, A.I., Akbulut, M., Çetin, A. (2017). Grain amino acid composition of barley (*Hordeum vulgare* L.) cultivars subjected to selenium doses. *Turkish Journal of Biochemistry*. 43(3), <https://doi.org/10.1515/tjb-2017-0027>