SELENIUM AND CHANGES OF AMINO ACIDS CONTENT IN GERMINATED BARLEY GRAINS

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Abstract
The present work studies the effect of selenium (Se) on amino acids content in barley after soaking grains in solutions with different sodium selenate concentrations.

Barley (Hordeum vulgare L.) grains were soaked in sodium selenate solutions with selenium concentration 1, 5, 10, 25, 50, 100 and 200 mg l$^{-1}$ for 5 days. Grains soaked in dionised water were used as control. Content of amino acids after germination was determined by Automatic Amino acid Analizator (AAA) T339 (Microtechna Praha).

Comparing content of separate amino acids after soaking grains in control solution and solutions with Se additives, we obtained that influence of Se depends on applied Se concentration. The highest increasing of amino acids content we observed regarding Valine – at Se concentration 50 mg l$^{-1}$ and Methionine – at Se concentration 5 mg l$^{-1}$. Se concentration 100 mg l$^{-1}$ gives increasing of 7 amino acids (Threonine, Serine, Glutamic acid, Glycine, Alanine, Leucine, Lysine). The highest applied Se concentration 200 mg l$^{-1}$ promotes increasing of content of Tyrosine and Phenilalanine.

Selenium additives promote increasing of content of all investigated amino acids, excepted Isoleucine.

We can conclude that selenium influences the forming of amino acids in barley grains during germination and it depends on Se concentration in solution.

Key words: selenium, barley, amino acids, germination.

Introduction
The barley (Hordeum vulgare L) grain, like other cereals, contains different chemical compounds–carbohydrates (78–83 g 100 g$^{-1}$) proteins (8-15 g 100 g$^{-1}$), lipids (2–3 g 100 g$^{-1}$), minerals (2–3 g 100 g$^{-1}$), and small quantities of B-group vitamins, including thiamine (B$_1$), riboflavin (B$_2$), nicotinic acid, and pyridoxine (B$_6$), as well as pantothenic acid, biotin, folic acid, and vitamin E (5–6 g 100 g$^{-1}$) (MacGregor, Fincler, 1993). These components provide valuable nutrients required by humans and also domestic animals.

Barley is known for its high content of dietary fiber, which has been shown to lower plasma cholesterol, reduce glycemic index, and reduce risk of colon cancer (Anderson et al., 1990; Jadhav et al., 1998, Slavin et al., 2000).

Barley contains all the known tocols (tocopherols and tocotrienols) (Peterson, 1994). The concentration of $\alpha$-D-tocotrienol, known as an inhibitor of cholesterol synthesis in liver of experimental animals, is higher in barley than in other grains. A second cholesterol inhibitor, $\alpha$-linoleic acid, has also been found among the barley fatty acids. The recently recognized involvement of antioxidant compounds in preventing the formation of carcinogens from precursor compounds has directed attention to such barley compounds as phenolic acids, phytin, vitamin E, proanthocyanidins, and catechins (Slavin et al., 2000).

Barley is also a good source of B-group vitamins, especially thiamine, pyridoxine, pantothenic acid, as well as biotin and folacin. Phosphorus, potassium, and calcium predominate among the barley mineral components (Encyclopedia of grain science, 2004).

Cereal grains provided 45% of our total protein intake. Genotype, environment, and growing conditions affect the amount of protein in kernel. Protein quality is mostly dictated by amino acid content and digestibility. The apparent protein digestibilities in cereals range from 80–90% (Encyclopedia of grain science, 2004).

The distribution of various chemical constituents is not uniform throughout the component tissues of barley grains. The husk and pericarp, the two outermost and protective tissues of barley grain, consist primarily of cellulose, hemicellulose, lignin and lignans. The embryo is rich in protein (34 g 100 g$^{-1}$), lipids (14–17 g 100 g$^{-1}$), ash (5–10 g 100 g$^{-1}$), sugars (sucrose 15 g 100 g$^{-1}$) (Encyclopedia of grain science, 2004).

For all cereals, the most limiting amino acid is lysine. The next most limiting amino acids are tryptophan and threonine.
Amino acids are used both metabolically, as building blocks for protein biosynthesis, and catabolically, as energy sources. Catabolism for most amino acids proceeds through transamination pathways; the exceptions are lysine and threonine. Specific enzymes catabolize these nutritionally limiting amino acids: threonine dehydratase acts on threonine and lysine ketoglutarate reductase on lysine (Shewry et al. 1994, Encyclopedia of Grain Science, 2004).

Although selenium (Se) is not an essential element for plants, it is an essential micronutrient for both humans and animals. More then 20 different selenoproteins have been characterized, including glutathione peroxidases and thioredoxin reductase, which are involved in controlling tissue concentrations of highly reactive oxygen-containing metabolites (Forduce, 2005). The consumption of food provides the principal route of Se intake for the general population.

Average Se concentration in cereals amounts 0.024 mg kg\(^{-1}\) of grain dry matter, ranging from 0.006 to 0.122 mg kg\(^{-1}\), for barley 0.06 mg kg\(^{-1}\), for wheat 0.011 mg kg\(^{-1}\) on the average (Morris, Levander, 1970).

Low dietary ingestion of Se has been assumed to contribute to an increased risk of cardiovascular disease and cancer and to promote infectious viral diseases related to heart disease and AIDS. It is known that human body should contain 5–20 mg of Se (Combs, 2001). The Latvian reference nutrient intake for Se is 60 µg day\(^{-1}\) (Ministry of Welfare, 2001).

In the present work, the process of sprouting was used to enrich selenium in barley Sprouting additionally improves the nutritional value of seeds, for example, by a higher vitamin content, a better quality of protein, and some other parameters (Lintschinger et al., 2000). Sprouting grains causes increased activities of hydrolytic enzymes, improvements in the contents of total proteins, fat, certain essential amino acids, total sugars, B-group vitamins, and a decrease in dry matter, starch and anti–nutrients. The increased contents of protein, fat, fibre and total ash are only apparent and attributable to the disappearance of starch.

However, improvements in amino acid composition, B-group vitamins, sugars, protein and starch digestibilities, and decrease in phytates and protease inhibitors are the metabolic effects of the sprouting process.

The present study investigates the influence of selenium additives on changes of amino acids content during barley grains germination using solutions with different selenium concentrations.

**Materials and Methods**

The research was performed at the Laboratories of the Department of Chemistry at the Latvia University of Agriculture and at the Laboratory of Biochemistry and Physiology of Animals at the Institute of Biology of University of Latvia.

The barley grains were germinated for 5 days. Grains (100 g) were soaked in 500 ml of solutions containing 1, 5, 10, 25, 50, 100 and 200 mg l\(^{-1}\) of selenium in form of selenate at ambient temperature of 18±2 °C for 6 h. The solution was then drained off and samples were germinated for 5 days under natural light conditions at 18±2 °C. Every 24 h the grains were moistened with corresponding solution and carefully shaken. Control sample without selenium additives was prepared for comparison of obtained results.

After germinating all grains, which were soaked in selenium-containing solutions, 3 times were washed with 500 ml deionized water to prevent contamination of the surface of grains with the solution containing selenium. After that, the grains were put into plastic packs and stored at –18 °C in a freezer for 24 h, then defrosted, dried and ground.

The content of amino acids was determined using AOAC Official Method 985.28 with Automatic Amino Acid Analyzator T339 (Microtechna Praha). The germination was performed in duplicate and analysis was carried out in triplicate. The data given here are the mean values of the measurements.
Results and Discussion
Sprouts are rich in digestible energy, bioavailable vitamins, minerals, amino acids, proteins, beneficial enzymes and phytochemicals, as these are necessary for a germinating plant to grow. These nutrients are essential for human health.
Nutritive value of proteins is determined by amino acid content, especially the quantity of essential amino acids. Laboratory studies have showed that selenium additives have influence regarding the content of essential amino acids.
The data of changes of essential amino acids content in barley grains after germination using solutions with different selenium concentrations is shown in Figure 1 and 2.

Figure 1. Content of essential amino acids at Se concentrations 1, 5 and 10 mg l\(^{-1}\)

Figure 2. Content of essential amino acids at Se concentrations 25, 50, 100 and 200 mg l\(^{-1}\)

Analysing obtained results we can see that selenium additives promote changes of amino acids content in barley grains, and it depends on Se concentration. For, example, Se concentration 5 mg l\(^{-1}\) promotes the highest increasing of Methionine (Met) content. Se concentration 50 mg l\(^{-1}\) promotes increasing of Valine (Val), but concentration 100 mg l\(^{-1}\) promotes increasing of Threonine (Thr), Leucine (Leu) and Lysine (Lys) contents in barley grains. As mentioned above, the most limiting amino acids for cereals are lysine and threonine. After 5 days germination using solution with selenium concentration 100 mg l\(^{-1}\) it is possible to increase the content of these amino acids: for 16.2% (Lys) and for 22.7% (Thr).
The highest applied Se concentration 200 mg l\(^{-1}\) promotes increasing of Phenilalanine (Phe) and Tyrosine (Tyr) contents. Different concentrations of Se promote changes of all essential amino acids content except Isoleucine (Ile).
Figure 3 and Figure 4 show the changes of non–essential amino acids content in barley grains after germination using solutions with different selenium concentrations.

![Graph showing content of non-essential amino acids](image1)

**Figure 3. Content of non-essential amino acids at Se concentrations 1, 5 and 10 mg l\(^{-1}\)**

![Graph showing content of non-essential amino acids](image2)

**Figure 4. Content of non–essential amino acids at Se concentrations 25, 50, 100 and 200 mg l\(^{-1}\)**

The obtained results are very similar regarding essential amino acids. As we can see, all applied Se concentrations promote increasing of investigated non–essential amino acids and it depends on Se concentration. For example, the one of the lowest applied Se concentrations – 5 mg l\(^{-1}\) promotes increasing of Aspartic acid (Asp), but Se concentration 10 mg l\(^{-1}\) promotes the highest increasing of Arginine (Arg). Se concentration 100 mg l\(^{-1}\) promotes the highest increasing of 4 non–essential amino acids: Serine (Ser), Glutamic acid (Glu), Glycine (Gly) and Alanine (Ala). Only the highest applied Se concentration 200 mg l\(^{-1}\) didn’t increased the content of investigated amino acids.

**Conclusions**

1. It is possible to change the content of amino acids in barley grains after 5 days germination using Se containing solutions.
2. Se additives promote increasing of content of all investigated amino acids except content of Isoleucine.
3. Se influences the forming of amino acids in barley grains during germination and it depends on Se concentration.
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References