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Assessment of the Impact of Selenium Fertilization on Nitrates (III) Content in Spinach (Spinacia Oleracea L.)

Ewelina Grochowska[1], Małgorzata Rosikiewicz-Wiankowska[2], Monika Komorowska[3], Marcin Niemiec[4]

[1, 2] Spółka Agrarna AR Sp. z o.o., Zdunowo 48, 09-142 Załuski, Poland

[3,4] Department of Agricultural and Environmental Chemistry, University of Agriculture in Krakow,21 Mickiewicza Av., 31-120 Krakow, Poland;

Abstract: Selenium (Se) is very important element for human health. It is involved in the body's defense mechanisms and hormone biosynthesis. Selenium is an element that occurs in trace amounts in the human body, is involved in the protection of cell membranes and has anti-cancer properties. Selenium is a cofactor of glutathione peroxidase and plays an important role in preventing oxidative tissue damage. The aim of the study was to assess the effect of selenium fertilization of spinach Spinacia oleracea L. on the content of nitrates(III) and nitrates(V) under interrupted refrigeration conditions. The experiment was carried out on spinach plants after foliar fertilization. The experimental factor was the form of introduced selenium. The experiment included 2 experimental objects and a control object, without selenium fertilization. Foliar fertilization was applied 14 days before harvest. After harvest, the plant samples were divided into two parts, one of them was preserved and the other was stored at room temperature for 72 hours. The content of nitrate III and nitrate V was determined. The test results confirmed the positive effect of selenium on the reduction of nitrate III content, with increased effectiveness for the used form of sodium selenate.

Keywords: biofortification, selenium, spinach, nitrate (III), nitrate (V), healthy food, and vegetables.

Introduction

Ensuring the safety of food products is a key element of all quality management systems. It is implemented based on legislative guidelines that are mandatory in the country of production and the not obligatory requirements, based on the optionally implemented and certified quality management systems, such as GLOBAL G.A.P (Good Agriculture Practice), SAI Platform (Sustainable Agriculture Initiative), QS (quality scheme for food).

Fresh vegetables are an important element of the human diet. They provide many valuable nutrients, including vitamins, minerals, fiber, but also substances with antioxidant properties (Kumar et al., 2020). The most important of them include polyphenols, carotenoids, selenium, zinc, vitamins A, C, E (Karmińska et al., 2017). As mentioned by Karmińska (2007), spinach is classified as a vegetable with high antioxidant activity. Spinach (*Spinacia oleracea* L.) is an annual plant belonging to the Amaranth family. The most common and widely used species of spinach is vegetable spinach (*Spinacia oleracea* L). Spinach can be eaten raw, e.g. as an ingredient in salads and cocktails, and can also be heat treated and used for stewing or baking. Spinach is also suitable for processing purposes, e.g. for freezing. Spinach, compared to other vegetables, has a high content of nutrients. 100 g of fresh spinach contains 235 mg of potassium, 53 mg of magnesium, 93 mg of calcium, 29 mg of phosphorus and β-carotene (4243 µg), vitamin A (707 mg), vitamin C (67.8 mg), vitamin E (1, 88 mg) (Kunachowicz et al., 2017).

The nutritional value of leafy vegetables, which should be a permanent component of a healthy diet, may be influenced by the availability of nutrients. The content of certain elements can be increased in the soil or nutrient solution to increase their concentration in edible vegetable leaves (Butri et al. 2021). This procedure is also called biofortification. This is a processes or treatments by which the nutritional value of plants grown for food purposes is improved through specific agronomic practices in order to improve human nutrition.

Selenium is a mineral involved in a number of biological functions in plants and animals. It is also a cofactor of important enzymes such as thioredoxin reductase and glutathione peroxidase, which protect against oxidative stress. Selenium is a component of enzymes such as selenocysteine in plants (Chao et al., 2022). There are also many other forms of organic selenium, such as selenomethionine, selenomethylselenocysteine, selenocysteine, selenomethylselenomethionine, dimethylselenopropionate, dimethylselenide, and dimethyldiselenide (Ellis et al., 2013). The main function of selenium is its antioxidant properties, protecting cells in plants, humans and animals (Fernández-Lázaro et al., 2020). The antioxidant capacity of selenium is very important in protecting the cell membrane and preventing cancer in humans (Pyrzynska, Sentkowska, 2021). However, the anticancer effect and its beneficial effects on human health are achieved in a very narrow range of selenium concentrations. Concentrations higher than the recommended daily intake may be hazardous to the health of consumers (Consentino et al., 2023).

The selenium content in edible plant parts depends on its concentration in the soil and the ability of plants to absorb it. Selenium available in the soil depends on its forms, microflora, biological, chemical and physical properties. In agricultural systems with low selenium concentrations, selenium can be fed directly by adding it to fertilizers. This strategy should be used with extra care, because a high selenium supply may cause accumulation in the soil, with the risk of excessive selenium uptake by plants. Biofortification of plants with selenium can also be achieved by foliar fertilization, avoiding accumulation in the soil (Boldrin et al., 2013).

Leaf plants are characterized by an increased risk of accumulating nitrates(III) and nitrates(V). This is a standard process, a consequence of nitrogen fertilization (Moćko, Wacławek 2005), although accumulation may also occur under conditions of proper agrotechnics. The content in food products also depends on the plant species and environmental factors. The largest amounts are found in young plants (Stasiak, Wilska-Jeszke J., 1983), so attention should be paid to the contamination of plants in baby stage (Rostkowski et al., 1994).

Nitrates (V) do not have a toxic effect on living organisms, however, as a result of the action of microorganisms, they are transformed into nitrates(III). Nitrates(III) are compounds with strong mutagenic, carcinogenic and teratogenic effects. The process of reducing nitrates(V) to nitrates(III) intensifies especially during the logistics process (Steiner et al., 1996). The factor that intensifies, among others, the process of nitrate(V) reduction is the disturbance of the refrigeration sequence.

The aim of the study was to assess the effect of selenium fertilization of spinach (*Spinacia oleracea L.)* on the content of nitrate(III) under interrupted refrigeration conditions.

Materials and methods

The aim of the study was achieved based on a vegetation experiment conducted in the period 12-26.10.2023, in the field located (52°29'46.6"N 20°31'00.9"E). The experimental factor was the form of selenium used for the fertilization. The experiment included 2 experimental objects and a control object, without selenium fertilization. The experimental objects were commercial foliar fertilizer containing selenium and sodium selenate solution. The dose of foliar fertilizer was 100 g/ha of pure selenium. Foliar fertilization was applied on 12.10.2023, 14 days before harvest. After harvesting, the samples were divided into two parts, one of them was preserved and the other was stored at room temperature for 72 hours. After this time, the content of nitrates(V) and nitrates(III) was tested. Nitrates(V) were determined by the colorimetric method using salicylic acid, while nitrates(III) were determined by the colorimetric method using sulphanilic acid. Additionally, the selenium content in the biomass was determined using the ICP OES method with inductively excited plasma.

Results and discussion

Table 1. Content of selenium and nitrates(III) and nitrates(V) in the biomass of freshly harvested plants

Experimental variant	Selenium content	Nitrate(V) content	Nitrate(III) content
	mg □ kg^{-1} fm		
Control object	Traces	4123	1,423
Commercial fertilizer	1,28	4354	1,322
Sodium selenate	3,56	4587	1,58

Table 2. Content of selenium and nitrates(III) and nitrates(V) in plant biomass after 72 hours of storage

Experimental variant	Selenium content	Nitrate(V) content	Nitrate(III) content
	mg □ kg^{-1} fm		
Control object	Traces	3855	52,85
Commercial fertilizer	1,45	4288	36,81
Sodium selenate	3,485	4321	33,85

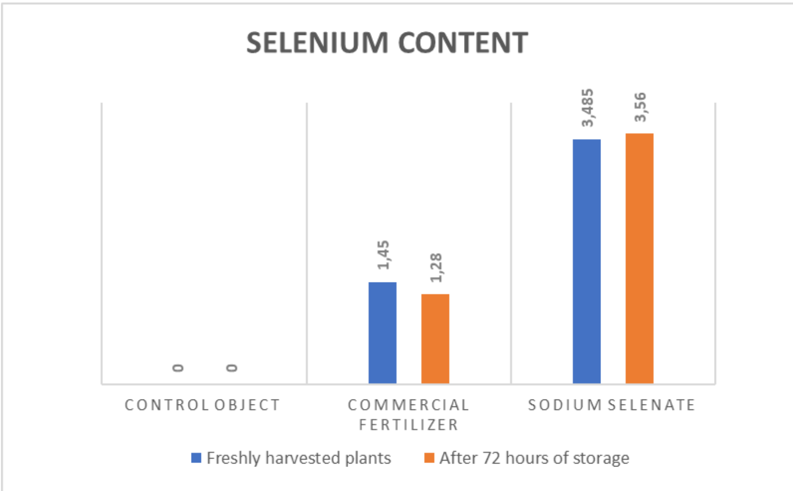

Figure 1. Content of selenium in different experimental variants

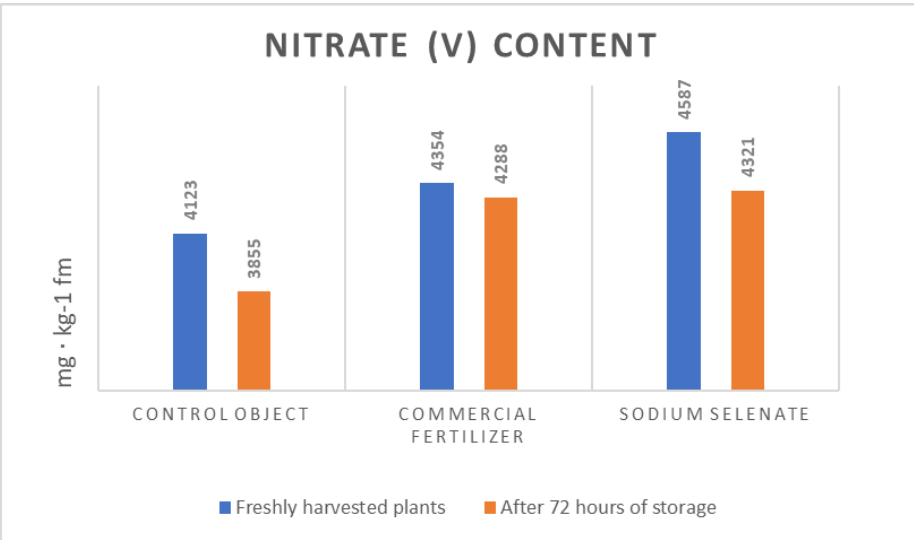

Figure 2. Content of nitrate(V) in different experimental variants

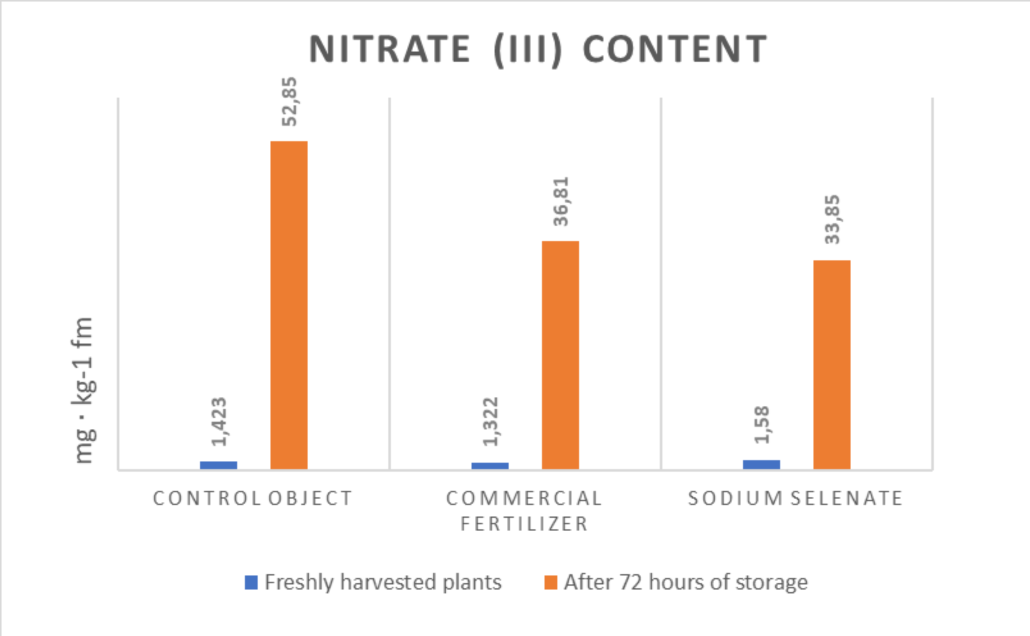

Figure 3. Content of nitrate(III) in different experimental variants

The experiment indicates differences in selenium content in tissues, depending on the fertilization type. Despite the use of the same doses of selenium, plants fertilized with a selenate solution, had a 270% higher selenium content after harvesting and a 240% higher selenium content after 72 hours of storage compared to samples fertilized with commercial fertilizer.

In all samples, a slight decrease in nitrate(V) content was observed after the storage period. The interruption of the cold chain indicated an increase in nitrate(III) content in all samples. The growth in samples treated with selenium was significantly lower than in the control sample. In the case of commercial fertilizer, a 25% reduction in nitrate(III) was recorded during storage, and in the case of sodium selenite by 42% compared to the control.

Results of research conducted by SHI Ya-jing et al. (2019) directly confirm that exogenous selenium has a positive effect in reducing the accumulation of nitrate(III) by increasing the activity of the nitrogen-metabolizing enzyme in spinach and lettuce. This study suggests that five μmol L–1 Se can be used to reduce nitrate(III) and increase lettuce and spinach yields. Simmilar resuls were obtains by Rioss JJ. and colleagues (2010),showed that the use of selenium at a dose of 10–120 μmol L-1 as selenite reduces the concentration of nitrates in lettuce.

Conclusion

This study confirms the positive effect of selenium on the reduction nitrate(III) content, with increased effectiveness for the used form of sodium selenate form.

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