The optimization of table beet cultivation by using a boron-containing chelate

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Abstract. In the field experiment, the possibility of optimizing the cultivation of table beet Beta vulgaris L. using a borate complex based on ethylenediamine disuccinic acid (B-EDDS) was investigated. Before sowing, beet seeds were soaked for a day in a solution of this compound, as well as in solutions of boric acid and a borate complex based on ethylenediaminetetraacetic acid (B-EDTA), taken for comparison, with a concentration of solutes of $1.5 \times 10^{-3}$ mol/L. During the growing season, the plants were sprayed twice with experimental solutions at a rate of 100 ml/m$^2$. In the first decade of August, the content of photosynthetic pigments in plant leaves and microbial carbon in the soil was determined, after harvesting, sugars and betanin in root crops were determined by spectrophotometric methods. The boron content in beet root crops was analyzed by the fluorometric method. According to the results of the two-year experiment, all three boron-containing compounds, to varying degrees, had a positive effect on the experimental plant. In decreasing efficiency, they can be arranged in a row: B-EDDS > H$_3$BO$_3$ > B-EDTA. Treatment with the solution of borate-ethylenediamine disuccinate complex increased the boron content in root crops by 65% in comparison with treatment with boric acid. In comparison with the control variant, the sugar content in root crops increased by 37%, the content of betanin increased by 25% and the yield of root crops increased by 39%. At the same time, the mass of microbial carbon in the soil increased by 20%, which serves as one of the arguments confirming the ecological safety of the compound under study. Judging by the results of the experiment, borate-ethylenediamine disuccinate has good prospects for use as boron micronutrient fertilizer.

1. Introduction
It is known that complexonates of trace elements-metals have been successfully used for several decades in various fields of practice, including plant growing [1–3]. Literature sources contain information on the chemical properties and some data on the biological activity of chelate compounds of macro- and microelements-metals based on common chelating agents: ethylenediaminetetraacetic acid (EDTA), iminodiacetic acid (IDA), nitrilotriacetic acid (NTA). The chemical industry, both Russian and foreign, produces complexonates of biometals for the needs of agriculture and complexes used in various industries, medicine, pharmacy, and food production. Among them, chelates based on EDTA are dominant [2-3]. Due to the rather high chemical stability of these compounds, which appear as a result of their use in the natural environment, a number of environmental problems arise [4, 5] caused by the accumulation of these substances in the soil and water of open water bodies.
One of the most promising options for resolving these problems may be the replacement of widely used effective chelating compounds with their environmentally friendly analogues, which are characterized by high chelating ability and undergo physicochemical and biological destruction when released into the environment. Complexone derivatives of dicarboxylic acids (CDDA) and, first of all, complexone derivatives of succinic acid (CDSA), as well as complexonates based on them, can be considered as compounds that meet these requirements [6].

Nonmetallic boron is absorbed from the soil by the root system of plants in the form of borate anions capable of complex formation [7]. Root vegetables, including table beet Beta vulgaris L., are most demanding on the boron content. This vegetable plant occupies one of the leading places among vegetable crops, which is explained by the high taste and dietary value of root crops. Boron deficiency causes a drop in the yield and quality of root crops. For this reason, the search for new effective and environmentally safe boron compounds suitable for use as micronutrient fertilizers seems to be a very urgent task.

In order to compare the effectiveness of various boron microfertilizers, traditional boric microfertilizer (boric acid, $\text{H}_3\text{BO}_3$) and chelated boron-containing compounds based on ethylenediaminetetraacetic acid (EDTA) and ethylene diamine disuccinic acid (EDDS)—respectively, B-EDTA and B-EDDS—in 2017–2018, a small-plot field experiment was performed.

2. Materials and methods

When performing the experiment, the following reagents and devices were used: $\text{H}_3\text{BO}_3$ (reagent grade), EDTA (reagent grade), $\text{H}_2\text{SO}_4$conc. (reagent grade), $\text{HCl}$conc. (reagent grade), NaOH (reagent grade), CuSO$_4$H$_2$O (chemically pure), quinalizarin. EDDS, B-EDDS and B-EDTA were synthesized by the authors according to well-known methods [6, 7]. The seeds of an experimental culture were: table beetroot of the "Krasniy Shar" variety ("Gavrish" LLC). The devices were: Spectrophotometer "SF-56", fluorimeter "Fluorat-02".

Judging by the value of exchangeable acidity ($\text{pH}_{\text{KCl}} = 5.9$) and the value of hydrolytic acidity ($\text{H}_8 = 2.15 \text{ mg-eq./100 g of soil}$), the soil of the experimental plot had a reaction close to neutral. The soil contains an average amount of humus (2.1%) and exchangeable potassium (110 mg/kg of soil) and a high amount of mobile phosphorus (216 mg/kg of soil). Among the cations of exchangeable bases, the amount of Ca$^{2+}$ prevailed (4.1 mg-eq./100 g of soil). The value of the degree of soil saturation with bases turned out to be quite high (86%). The boron content in the soil was 0.28 mg/kg of soil.

The area of the experimental site was 200 m$^2$; the area of the accounting plot was 5 m$^2$, the repetition of the variants was fourfold, their location was randomized.

Table beet seeds, pre-soaked for 24 hours in test solutions with a concentration of dissolved substances of 1.5·10$^{-3}$ mol/l, were sown in open ground at the beginning of June according to the experimental scheme. Seeds soaked in $\text{H}_2\text{O}$deion. served as control. No macrofertilizers were used in the experiment.

After the appearance of the third true leaf, the plants were sprayed with the test solutions with a concentration of 1.5·10$^{-3}$ mol/L at a rate of 100 ml/m$^2$. Spraying was repeated after 10 days.

In the first ten days of August, samples of beet leaves were taken to determine the content of photosynthetic pigments [8] and soil samples were taken to determine microbial carbon [9]. After harvesting, the boron content in root crops was determined by the fluorometric method [10], and sugars and betanin [11] in root crops were determined by the spectrophotometric method.

3. Results and Discussion

The analysis data are presented in the diagrams (Figs. 1-6). Evidently from the diagram in Fig. 1, boron is most efficiently absorbed by beet plants in the form of a borate-ethylenediamine disuccinate complex, similar to chelated forms of biometals [1-3].

Since boron is involved in the synthesis of chlorophyll [12], an increase in its content in a plant has a favorable effect on the growth of the level of green and yellow pigments (Fig. 2) and, consequently, on the intensification of the processes of photosynthesis.
In addition to chlorophyll synthesis, important functions of boron in plants are participation in the formation and transport of carbohydrates, in particular, boron intensifies the formation of sucrose disaccharide from glucose and fructose. The dependence of the content of sugars, presented in beet root crops mainly by sucrose, on the level of boron compounds was confirmed by the results of the study: the minimum sugar content (7.3%) was found in the root crops of control plants, the maximum (10%) was found in the 4th variant of the experiment when using B-EDDS.

Boron is also involved in the synthesis of phenolic compounds [12]. These include betanin. Betanin is a red-violet compound that largely determines the dietary value of beetroot, has a high antioxidant activity, prevents atherosclerosis, prevents the occurrence of malignant tumors, and dysfunctions of the gastrointestinal tract.

Boric acid and B-EDDS to the same extent contribute to an increase in the content of this substance in root crops (Fig. 3).

Apparently, the ethylenediamine disuccinate ligand does not take any part in the metabolism of phenolic derivatives. When using B-EDTA, the betanin content in root crops is slightly reduced. The copper-containing enzyme phenol oxidase is involved in the initial stage of betanin synthesis [11]. It can be assumed that B-EDTA, penetrating into the plant cell, is involved in ligand exchange reactions and inactivates phenol oxidase, forming a more stable chelate complex with copper (II). Due to these changes, the accumulation of betanin is slowed down. At the same time, borate anions are released, which have some positive effect on plant metabolism.
When foliar treatment of vegetative plants with solutions, part of them inevitably enters the soil and has one or another effect on the soil microflora and biota. The nature of the effect on the microflora can be judged by the level of microbial carbon content.

![Figure 3](image3.png)

**Figure 3.** Change in betanin content (% of control) in beet root crops when treating with solutions: 1) control, 2) H₃BO₃, 3) B-EDTA, 4) B-EDDS (the content of betanin in the roots of control plants was 556 mg/100 g of raw material).

Although boron is not considered a necessary trace element for most microorganisms [12], foliar feeding with boron-containing substances in the presented experiment caused a noticeable increase in microbial biomass (Fig. 4), which can serve as an additional argument for the environmental safety of the compound under study.

![Figure 4](image4.png)

**Figure 4.** Change in the content of microbial carbon in the soil (% of control) after treatment with solutions: 1) control, 2) H₃BO₃, 3) B-EDTA, 4) B-EDDS (the content of microbial carbon in the soil of the control plots was 67 μg/g).

The processing of table beets with solutions of all boron-containing compounds made it possible to increase the yield of root crops to varying degrees (Fig. 5). The maximum yield was established when using B-EDDS.

In decreasing efficiency, boron-containing compounds can be arranged in a row: B-EDDS > H₃BO₃ > B-EDTA. It should be assumed that the plant uses not only boron, but also an organic ligand as an additional source of nitrogen and organic carbon from the B-EDDS chelate complex. Even if the ethylenediaminetetraacetate ligand is used by a plant, it is very limited due to its low biodegradability [4].
Figure 5. Change in the yield of table beets (% of control) as a result of processing by solutions: 1) control, 2) H$_3$BO$_3$, 3) B-EDTA, 4) B-EDDS (yield of root crops on control plots was 5.0 kg/m$^2$).

4. Conclusion
The results obtained over 2 years of research showed the highest efficiency and environmental safety of the use of the borate complex B-EDDS in comparison with the traditional boric micronutrient fertilizer—boric acid and the borate complex of ethylenediaminetetraacetic acid, B-EDTA. The chelate complex B-EDDS can be recommended for practical use as a new environmentally friendly boron microfertilizer that stimulates not only an increase in yield, but also an improvement in the quality of beet root crops.

References
[1] Dyatlova N M, Temkina V Ya, K.I. Popov K I 1988 Complexones and complexonates of metals (Moscow: Chemistry) 544 p
[2] Ostrovskaya L K 1986 Complexones and complexonates of metals and their importance for plant nutrition with trace metals Physiology and biochemistry of cultivated plants 18(6) 591 – 603
[3] Bitutsky N P, Kashchenko A S, Kozhev V P 1991 The effect of chelating agents and chelating agents on the chemical composition of plants Agrochemistry 10 99-107
[4] Dedyukhina E G, Chistyakova T I, Minkevich I K 2007 Biodegradation of EDTA Bulletin of biotechnology and physical and chemical biology 3(2) 40-49
[5] Caparullina E N, Fedorov D N, Doronin N V, Trotsenko Yu A 2008 A bacterial strain characterized by the need for EDTA Applied Biochemistry and Microbiology 44(4) 399–403
[6] Smirnova T I, Khizhnyak S D, Nikol'skii V M, Khalyapina Y M, Pakhomov P M 2017 Degradation of complexons derived from succinic acid under uv radiation Russian Journal of Applied Chemistry 90(4) 406-411. DOI: 10.1134 / S1070427217040024.
[7] Tolkacheva L N 2012 Physicochemical study of the processes of complexation of subgroup III-A elements with complexone derivatives of succinic acid (Tver, 2012) pp 57-60
[8] Gavrilenko V F, Zhigalov T V 2009 Big workshop on photosynthesis (Moscow: ACADEMA) pp 46–63
[9] Ganzhara N F (ed) 2002 Workshop on soil science (Moscow: Agroconsult) pp 108-109
[10] Saenko I I, Tarasenko O V, Deineka V I, Deineka L A 2012 Betacyanins of red table beet Scientific Bulletin of Belgorod State University. Natural sciences 3 194-200
[11] Pashkevich E B, Suvorov E E, Verkhovtseva N V 2011 Physiological and biochemical functions of boron in a plant Agrochemistry 11 85-96