

**COMPARATIVE RESEARCH CHANGE OF DYNAMICS AMOUNT
OF SUGARS IN THE LEAVES OF DEPENDENCE OF HIGH SALT
CONCENTRATION OF SOME VARIETIES OF SUGAR BEET (BETA
VULGARIS L.)**

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Abstract

In this article investigated comparative biometric indicators and functional status of stomata in leaves, changes in the dynamics of sugar levels at high salt concentrations of NaCl and Na₂SO₄ in leaves of sugar beet varieties (Beta vulgaris L.) Cooper, Taltos, Tarifa. It was revealed that the stress of salts in all three varieties of sugar beet leads to the negative impact of a change in morpho - physiological parameters. According to the parameters studied, it was established that, in comparison with the effects of salt stress, the Tarifa variety is more salt-resistant than the Taltos and Cooper varieties.

Keywords: sugar beet, salt stress, biometric indicators, stomata, adaptation

Introduction

Plant development depends on external factors, such as light, temperature, humidity of saline soils. Regulation of the course of biological processes occurs within these factors, and as a result, plant organisms in the lower and upper temperatures, drought, salinity, various diseases lead to the emergence of opposition. (Kasumov, 2018). Currently, 25% of the continent in the world is to some extent saline (Hassan et al. 2011), which has resulted in a loss of plant yields, a weakening of biodiversity and, at the end, an economic loss. (Bayat et al. 2010; Shevyakova et al. 2009). Considering the above mentioned for obtaining highly productive varieties of new cultivated plants, as well as adaptation to adverse conditions for scientists and researchers involved in breeding and in the agricultural sector one of the main tasks. Based on this context, it is possible to confirm that sugar beet does not lag behind grain crops both from its field of application and economic profitability. Therefore, a very important role is played and has a fundamental importance, the high scientific



and practical importance of research in the field of stress. The conditions of sugar beet are metabolic, biochemical, biometric and morpho-physiological concerning the organs of this technical plant.

Despite the fact that in the process of evolution in the organisms of various living organs, the influence of external factors on the path of adaptation occurs differently. It is necessary to recognize the fact that to cognize the above-mentioned factors and to defend against them, the protection mechanism has not yet been fully revealed [Elanskaya, 2005; Yelanskaya, Karandashova, 2006]. The response against the stress of a plant directly depends on the genotype, the duration of exposure to stress, the stage of its development, and the type of cell and tissue. Plant endurance is also determined by the degree of genome activity, the intensity of gene expression, the presence of joint genes [Akhundova, 2004]. From this point of view, identifying the mechanism of tolerance, increasing plant resistance to stress, first of all it is necessary to determine the physical, biochemical and genetic changes occurring in plants. This provides, in the sense that in this approach it is possible to determine their resistance and its definition playing an important role in individual genotypes to stress [Wangetal., 2003].

In the generation of stress resistance in plants, malate cells accumulated in plant vacuoles, organic acids like oxaloacetate, are of particular importance. A large number, of amino acids, including, proline, sugars, mineral salts, in the form of turgorogenous substances. It has been revealed that sugar and other carbohydrates, the accumulation of their heterotrophic plant tissues, plays an important role donor - acceptor bonds on the basis of the distribution of photoacciator. As a result, a concentration gradient is formed due to organic acids carried out through plant tissues. The driving force created by this method as a result of a physicochemical compromise in the reserve tissues, the accumulation of metabolites and their movement is realized. (Ho. 1988).

In the reserve cells of plants osmoregulated sugars vacuoles in the process of accumulation inside the vacuoles with the effect of acid inverses, the sugars are separated into glucose and fructose (Guern et.al., 1987). This enzyme in the active phase of numerous plant tissues reversible - the opposite process leads to a correlation of sucrose concentration (Yelleet.al., 1991). It also regulates the main tissues in reserve tissues as actively osmotic components of cell turgor generation. [Moore, Cosgrove, 1991]. As it is known sugar beet shoots are obtained in beets are obtained within 4-6 days. Secondary leaves are formed in 8-10 days. In sugar beet between ordinary leaves, the phase of their coverage, the development of plants occurs in 50-60th days. The plants studied by us are investigated before and after the indicated phases. Of the varieties Cooper, fast ripening, Tarifa u Taltos is medium ripening. The main objective of the study is to determine the comparative, biometric indicators in the varieties of sugar beet Cooper, Taltos and Tarifa, their leaves in normal and salt stress quantities, the distribution of stomata changes the dynamics of sugar.

Materials and methods

The object of the study is the exported from Holland sugar beet varieties Taltos, Cooper and Tarifa leaves of these varieties.

The seeds of sugar beet are placed in the vegetation vessels. After receiving the primary seedlings, the objects of the subsequent stage of investigation were brought



to 23-25 ° C temperature of the vegetation vessels, the photoperiod was 14 hours, the humidity was 60-70% and the intensity was 10-15 klux. Conducted research experience, artificially created phytotronchambers. In each control variant, irrigation was carried out until the end of the growing season; from the experimental variants, were cultivated in 1%, 2% and 3% concentration NaCl to one, another were cultivated 1%, 2% and 3% concentration Na₂SO₄. This created artificial salinization.

After 30, 45, and 60 days of periodic ontogenetic development of plants for about 12:00 hours, 3-4 hours of clarification separated the leaf samples.

The estuaries in the leaves of the created conditions, the situation under stress is an important biometric parameter that affects the intensity of transpiration and assimilation of CO₂. Therefore, in plants under normal and stressful conditions, the position in the stomata leaves and the results obtained on this basis play an important role in resistance against stressors. In the experiments, the position of the stomata is determined by a simple microscopic method, cutting (Lobomed lx400, USAP). The functional position of the stomata is determined in advance by the preparation of hypotonic, hypertonic, microscopic preparations or the observation of plasmolysis and deplasmolysis processes by the presence of water. The processes of plasmolysis and deplasmolysis were achieved with NaCl solution. This indicator was determined in the period of open and closed positions of stomata in the first stage. To do this, in a sunny day of a certain time (between 11: 00-13: 00), 2 leaves were taken from the stems of the middle part of the plant, a layer was removed from the reverse, the epidermis, one drop of diluted glue was added to the layers, then added to the plate.

It should be noted that, given the large number of stomata samples for the preservation of stable positions at the stage, it was introduced by a fixative (3: 1, acetic acid as ethanol). After that, the plates were transferred to the refrigerator and alternately opened and the stomata increased 400 times and the microscopic measurement was determined for each plate was carried out at least 3-5 times. If the size of the stomata opening was fixed below 0.004 mm, it was considered closed to the stomata. To obtain samples of leaf extract after thorough washing and drying with filter paper, the leaves were weighed. 1 gram of leaf was added with 7 ml of buffer - a solution consisting of 80mM, pH 7.4 phosphate buffer, 2 mM dithiothritol (DTT), 1mM etilendiamin acid tetra-acetate - 4Na (EDTA), 10 mM MgCl₂, 0.5 polyvinylpyrrolidone (PVP) u 1 %, Triton X-100), quartz sand was added at a temperature of + 4 ° C in a mortar was homogenized. The resulting homogenate was strained with double gauze. Then, for purification from residues of unfolded vegetable impurities, first for 300 g 15 min, after 2000 g 20 min. Spent precipitation. After separation of the precipitate with the upper liquid part, the studies were continued.

From the biometric parameters of sugar beet leaves, the leaf area, mass, width, length, light reflection, thickness were determined. To measure the area of the leaves (S_l), sugar beet revenge was kept for one hour between the leaves of the book. Then, placing on graph paper, the projection of the leaves was obtained with a thin sharp pencil in all respects. After removing the leaves, the projected area of the leaves is plotted on graph paper. All the biometric measurements associated with the mass of the test sheets - the width, length, ordinary ruler, and the weight of the leaves, roots, all the mass of the plant held ordinary weight.



Dry leaf weight (M_d) also applies to biometric parameters. To determine the M_d , the leaves first need to be isolated from the wood. Then, you need to be washed with distilled water, then dried with, filtered paper. After that, you need to dry until getting absolute dry mass. To obtain a dry mass should be dried at a stable 70°C temperature for 48 hours. At the end of the experiment, the dried leaves were weighed with an analytical balance.

The relative amount of water (moisture) SNM and water deficiency (H.B) are determined by the method of Tambussi [Tambussi et.al 2005]. To determine the total amount of water, it was first weighed in an analytical weight for 24 hours, kept under wet filtered papers. After, the leaves at 60°C were dried until completely dry. After complete saturation of the leaves with water, dry filtered paper isolated water droplets of the surface of the leaves, then weighed the leaves of the plant again. Then the samples were again thermostatic at 105°C until an absolute dry mass was obtained. What led to complete, drying. The calculation was carried out according to the following formulas.

Humidity = (1) SNM = (2)

SQ = (3)

Where M_f is the mass not saturated with water, M_t is the mass after saturation with water, M_d is the mass of dry leaves.

The amounts of chlorophylls in plants (a, b) were determined using the Sims u Gamon method (Sims, Gomon, 2002) with an 80% acetone extract in the leaves (acetone, Tris-HCl buffer Ph 7.8 = 80:20 volume).

Extract the leaves in the above composition indicated by a spectrophotometric method.

Determination of sugars in plant organs.

The amount of sugars in sugar beet leaves was determined by the Bertrand method [Ermakova, 2005]. This method is based on sugars containing a carbonyl group in a molecule that has reducing properties. These sugars of copper sulfate ($\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$) in an alkaline medium turns from the oxidized form into the reducing one. Which is accompanied by the action of aldose transformation of $\text{Cu}^{2+} + \text{Cu}^+$, otherwise copper ions are reduced. To determine the amount of sugars, acid or enzymatic hydrolysis of sucrose and other oligosaccharides is first required. Here the main task is a clear definition (calculation) of the amount of precipitated $\text{Cu}(\text{OH})_2$ and the corresponding soluble amount of sugars. The process is clearly indicated in the literature [Ermakova, 2005]. By the method under study, the amount of glucose should not exceed 10-90 mg and in the process should not be present amine salts, amino acids, as well as pectin's. They turn a precipitate of copper oxide into a dissolved state. The Bertrand method also measures glucose, fructose, galaxies, mannose, and lactose. In determining the sugars from the materials studied, it is also necessary to separate proteins. To this end, salts, heavy metals such as mercury, zinc hydroxide, Bernstein's solution of potassium hydroxide are added to the solution.

The total amount of dissolved protein was measured by a spectroscopic method at a wavelength of 750 nm. To draw up the calibration curve, albumin serum of large-horned ramp was used [Lowry, 1951].

The statistical analysis of the results obtained, the tables in the figures, the arithmetic average figures, reflected the quadratic deviations. In the analysis of completed research works, the arithmetic mean assumption and the deviation ($M \pm$



m) are taken into account. The estimates obtained are not less than three results of the repetition of biological and analytical studies. Standard errors and deviations in their calculation are made in the program Microsoft Office Excl.

The results and discussion

One of the sections of this research to induce 3 varieties of sugar beet in saline soils tied

From, Danni. Choose varieties associated biometric indicators. Some of these indicators (parameters), obtained shoots were carried out visually according to morphological features, the other part was determined by mathematical-geometric methods. The results obtained are listed in Table 1.

As can be seen from the table, the length of leaves and roots between varieties is observed regularly - proportionally. This result between the leaves and the root is more clearly (clearly) observed in their ratios (L_l / L_r). If you carefully look at the table, you will easily be convinced that the 3% Na_2SO_4 percentage effect is more stimulated by all varieties over a period of time. Therefore, in these conditions, in the leaves and in the roots of these varieties in the course of stimulation, the development of the root system in the organs closer to the development is optimal. Similarly, the results obtained are parallel to other biometric parameters. These signs are also observed in the leaves, roots, respectively, in the masses, and in the indicator area of the leaves.

The staff member discovered that in the presence of 50Mm NaCl the wet and dry weight of leaves and the shoot in *Theiungiellahalophila* plants and their number does not change, the membrane conductivity at the roots decreases, in the leaves and roots also small maladigehid (MDA) and active oxygen (AK) increases, the amount superoxide dismutase (SOD). In 200-400 mMNaCl – oh concentration of plants, its weight decreases, shoots and roots. At the same time increases and MDA and AK. In all concentrations of NaCl, the leaves increase membrane conductivity and catalase and peroxidase activity [Liu et al., 2006].

The impact of stress in the metabolism of plants occurring change in the first place is reflected in their morphological appearances and in the change in the dynamics of biometric indicators. Since the onset of stress and the passage of time, changes occur in their observation, fixation and analysis play an important role in the processes occurring in the cell and tissues, as well as metabolic transformations corresponding to their characters. According to this, as a sign of adaptation, the effect of salt in the plant root system in cells accumulates a large number of salts. They, increasing osmosis in the cells, prevent the release of water from root cells. Changes in plant organs, in salt stress, including leaf development, changes in architecture, from the development phase (ontogenesis) of stress levels, the type of weather conditions and the mineral composition of the soil. Biometric changes in plant organs are one of the main indicators in their resistance.



Table 1. Leaf and root length in sugar beet

kinds	Variety	L_y , mm	L_k , mm	L_y/L_k
30 day				
Cooper	Control	40	20	2
	2%NaCl	25	15	1,7
	3%NaCl	15	1	1,5
	2%Na ₂ SO ₄	37	23	1,6
	3%Na ₂ SO ₄	18	9	2
Taltos	Control	48	22	2,2
	2%NaCl	32	24	1,3
	3%NaCl	27	15	1,8
	2%Na ₂ SO ₄	29	21	1,3
	3%Na ₂ SO ₄	24	16	1,5
Tarifa	Control	50	25	2
	2%NaCl	34	21	1,6
	3%NaCl	28	18	1,5
	2%Na ₂ SO ₄	37	27	1,4
	3%Na ₂ SO ₄	35	20	1,7
45 day				
Cooper	Control	50	30	1,7
	2%NaCl	30	20	1,5
	3%NaCl	18	15	1,2
	2%Na ₂ SO ₄	42	27	1,6
	3%Na ₂ SO ₄	25	12	2,0



	Control	51	27	1,9
	2%NaCl	39	26	1,5
	3%NaCl	30	25	1,2
Taltos	2%Na ₂ SO ₄	32	24	1,3
	3%Na ₂ SO ₄	26	19	1,4
	Control	61	30	2,0
	2%NaCl	38	25	1,5
Tarifa	3%NaCl	30	21	1,4
	2%Na ₂ SO ₄	45	32	1,4
	3%Na ₂ SO ₄	47	24	2,0
60 day				
	Control	59	40	1,5
	2%NaCl	35	24	1,4
Cooper	3%NaCl	23	19	1,2
	2%Na ₂ SO ₄	48	32	1,5
	3%Na ₂ SO ₄	29	17	1,7
	Control	61	32	1,9
	2%NaCl	47	28	1,7
Taltos	3%NaCl	43	30	1,4
	2%Na ₂ SO ₄	38	29	1,3
	3%Na ₂ SO ₄	28	21	1,3
	Control	75	42	1,8
	2%NaCl	44	31	1,4
Tarifa	3%NaCl	32	23	1,4
	2%Na ₂ SO ₄	51	43	1,2
	3%Na ₂ SO ₄	52	27	1,9



As can be seen from table 2, in all three samples in the leaves of sugar, the amount of sugars varies insignificant amounts depending on the type (composition) and salt concentration. At a closer look, by reviewing the table, you can make sure that in all three varieties of sugar beet, the amount of sugars under the action of salts of NaCl and Na₂SO₄ in 2 and 3% concentrations increases in plants to 45 days old and develops gradually. In subsequent stages of ontogenesis, this indicator begins to gradually decrease. As expected, in the variety Tarifa compared with other varieties in all 3 stages, the amount of sugar increased more than 2 times. This pattern was taken into account in previous varieties. In the indicated table 2 options and the figures confirm that the amount of soluble sugars accumulated more in the Tarifa variety in the 3% Na₂SO₄ variant, the highest in the Cooper variety in the 2% -NaCl variety

If we look at the analysis of the amount of sugars by the type of salt solutions, we can be sure that the salted sulphates of 1 soil leaves the plants compared to chlorine saline soils of sulfate soils leaves compared to have an advantage. Here, only Tarifa variety has properties. In contrast to Cooper and Toltos varieties, in Tarifa variety, in both salt concentrations, the effect of NaCl and Na₂SO₄ salts is the dissolved amount of sugars in the leaves is approximately the same. This relative tendency has given Saib to encourage the subjects to be developed in all stages. It can be seen from the table that NaCl in control samples of each 2 concentrations of Na₂SO₄ differs in comparison with the increase in sugar content. This fact makes it possible to confirm that, for the selection of varieties exposed to salt stress, it is necessary to investigate (apply) not one but several salts. In our study, the variety Tarifa meets this requirement. As in comparison with the varieties of Cooper and Toltos, we found that the variety Tarifa is a more salt tolerant variety. The amount of sugars and localization in the leaves in salt-resistant varieties is compatible with photosynthesis. And the amount of pigment can provide us with even more information in which seedlings and some biometric indicators will be obtained, with the results studied. For this purpose, we studied the percentage of seedlings obtained and its intensity in the specified varieties of sugar beet. We have revealed that there is an accumulation of sugars in the leaves of sugar beet, its amount between the percentages of seedlings obtained there is a directly proportional ratio. The varieties we study Tarifa related more salt tolerant, as well as the advantages of similarity in percent. Apparently, the results we obtained are connected with the peculiarities of the genotype of the variety. That is, in resistance to salt effects also leads to high shoots.



. Analysis of amount of sugar in leaves of sugar beet (*Beta vulgaris* L) Table 2

kinds	variety	soluble carbohydrates/mg/ml		
		30 day	45 day	60 day
Cooper	control	5,3±0,20	6,2±0,23	3,5±0,18
	2% NaCl	4,5±0,21	5,7±0,14	3,9±0,19
	3% NaCl	8,3±0,28	9,4±0,29	6,6±0,33
	2% Na ₂ SO ₄	7,5±0,32	8,5±0,27	6,1±0,31
	3% Na ₂ SO ₄	14,7±0,51	16,2±0,63	10,3±0,51
Taltos	control	7,2±0,27	8,5±0,29	5,2±0,26
	2% NaCl	5,5±0,11	6,3±0,15	3,2±0,16
	3% NaCl	8,2±0,33	6,7±0,27	7,3±0,37
	2% Na ₂ SO ₄	14,5±0,51	15,7±0,55	12,1±0,61
	3% Na ₂ SO ₄	13,7±0,55	14,3±0,57	11,6±0,58
Tarifa	control	8,7±0,37	10,3±0,24	7,9±0,39
	2% NaCl	11,8±0,54	14,1±0,53	10,4±0,52
	3% NaCl	14,7±0,65	17,3±0,60	13,9±0,69
	2% Na ₂ SO ₄	14,4±0,68	16,1±0,57	13,5±0,67
	3% Na ₂ SO ₄	17,5±0,83	18,4±0,79	16,4±0,82

If we look at the analysis of the amount of sugars by the type of salt solutions, we can be sure that the salted sulphates of 1 soil leaves the plants compared to chlorine saline soils of sulfate soils leaves compared to have an advantage. Here, only Tarifa variety has properties. In contrast to Coopera and Toltos varieties, in Tarifa variety, in both salt concentrations, the effect of NaCl and Na₂SO₄ salts is the amount of sugars in leaves is approximately the same. This relative tendency has given Saib to encourage the subjects to be developed in all stages. It can be seen from the table that NaCl in control samples of each 2 concentrations of Na₂SO₄ differs in comparison with the increase in sugar content. This fact makes it possible to confirm that, for the selection of varieties exposed to salt stress, it is necessary to investigate (apply) not one but several salts. In our study, the variety Tarifa meets this requirement. As in comparison with the varieties of Coopera and Toltos, we



found that the variety Tarifa is a more salt tolerant variety. The amount of sugars and localization in the leaves in salt-resistant varieties is compatible with photosynthesis. And the amount of pigment can provide us with even more information in which seedlings and some biometric indicators will be obtained, with the results studied. For this purpose, we studied the percentage of seedlings obtained and its intensity in the specified varieties of sugar beet. We have revealed that there is an accumulation of sugars in the leaves of sugar beet, its amount between the percentage of seedlings obtained there is a directly proportional ratio. The varieties we study Tarifa related more salt tolerant, as well as the advantages of similarity in percent. Apparently, the results we obtained are related to the characteristics of the genotype of the variety. That is, resistance to salt effects also leads to high shoots.

Optimal experimental conditions were created to determine the percentage of germination and energy of germination, of each of the varieties taken from sugar beet. For this purpose, the mineral composition, temperature and soil moisture in vegetable containers were leveled to ensure sowing and seed germination, and 20 seeds were planted for each sample. Starting from the fourth day, the first shoots began to appear, on the sixth day the shoots were completed, and after this day new shoots were not observed. If we look at the table, we see that the percentage of sowing and seed germination in Tarifa is higher than that of the Cooper and Taltos varieties. Second place - Cooper, and the last place - Taltos

Energy and percentage germination sugar beet seeds

Table 3.

kinds	variety	seed quantities	germination energy	germination percentage
Cooper	Kontrol	20	20	80
	2%NaCl	20	15	75
	3%NaCl	20	12	60
	2%Na ₂ SO ₄	20	18	75
	3%Na ₂ SO ₄	20	16	60
Taltos	Kontrol	20	19	95
	2%NaCl	20	12	60
	3%NaCl	20	10	50
	2%Na ₂ SO ₄	20	14	70
	3%Na ₂ SO ₄	20	11	55



Tarifa	Kontrol	20	15	100
	2%NaCl	20	13	75
	3%NaCl	20	10	60
	2%Na ₂ SO ₄	20	15	90
	3%Na ₂ SO ₄	20	12	80

One of the diagnostic methods of researching plants to aridity and salt stress is under their influence. Under the influence of salt stress, as a result, in the leaves of plants, partial or stomata closing occurs, which leads to abnormal photosynthesis, does not allow passage of photosynthesis. The normal origin of photosynthesis is directly dependent on photosynthetic pigments. On this basis, we studied the effects of salt stress, the amount of chlorophyll (a, b, a + b, a / c) and the change in the processes taking place. The ratio of chlorophyll a / b in Tarifa leaves has advantages in comparison with others. It was revealed that Na₂SO₄ at a concentration of 200 mM Na₂SO₄, compared with other stressful effects, more highly stimulates the plant. According to the data obtained in the Tarifa in terms of salt stress, the amount of carotenoids is also high in comparison with other varieties.

By less the majority of scientists in the vacuoles of plant tissues in majesty of sugar and amino acids, Poling leads to an increase in turgor pressure in plant cells. This leads on the one hand to prevent the loss of water molecules, and on the other hand, to refrain abilities in the cell sap of water. And osmotic regulation takes place in the village of transpiration, continued the process of photosynthesis. There is such a mesh that, biosynthesis of turgogenic substances, their biosynthesis or the formation of these upon decomposition of macromolecules, requires energy consumption of energy, which is associated with consumption [Christopher, Tony, 2008; Riccardiet al., 2000]

Considering the above mentioned, it can be concluded that in the saline lands it is advisable to mix the Tarifa variety for sowing sugar beet. Since this variety, by the amount of sugars and pigments in the leaves, percentage yield and some biometric indicators, has advantages in comparison with other varieties.

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