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Influence of various salts of Na-isocated salts on the change in the activity of major NADPH-forming enzymes due to the development of bean seeds

B.G. ASADOVA
Azerbaijan State Pedagogical University, Baku
Azerbaijan

Abstract

As is known, most plants are in sedentary lifestyle. Therefore, they are often forced to be exposed to environmental extreme factors. One of the most important factors in these factors and in nature is the salinity of the soil, ie salt stress. The salinity factor often coincides with the drought factor and causes the negative effects of saline on the plants. The occurrence of such severe stress leads to the slowdown in growth and development of plants, the weakening of vitality, the sharp decrease in their productivity, and the destruction of plants when serious stress is present.

Key words: solutions of Na salts, NADPH-forming enzymes, bean sprouts

INTRODUCTION

The adaptation of plants to the environment, including environmentally unfavorable conditions, is accompanied by changes in metabolism, and NADPH is required to make these changes. The cell has four known enzymes (Q6PDH, 6PQDH, SYSDH and DMDH) that generate NADPH pulse, and the bases are Q6PDH and DMDH enzymes. With regard to salt stress, relative Q6PDH is learned from these two enzymes and the weakest is DMDH enzyme.

Q6PDH and the pentosophosphate pathway involved in its regulation are commonly occurring processes in nature, all primarily (excluding viruses) and higher organisms. The universality of ferment from the point of view of the spread of living things is apparently related to the importance of the function it performs in metabolism. Therefore, Q6PDH is a protein that has not been disturbed by researchers and actually a relatively well-known enzyme.

The localization of the enzymes of pentozophosphate in the oxidation of glucose in plant cells is strange. Enzymes that catalyze the cyclization of the oxidative and reducing phase were all found in plastids, including chloroplasts. Despite the discovery of all the fermentation of the oxidizing phase in the cytoplasmic fraction, it has not yet been possible to detect a number of fermentations of the reducing sequence. Therefore, it is believed that the pentozophosphate road operates in a reduced (reduced) form of cytosol. This allows the pentozophosphate to say that the Q6PDH enzyme that regulates the route, including its activity, serves different purposes (different purposes) in different cells of plant cells.

NADPH is required in the synthesis of the redundant form of glucose, one of the vital metabolites of the cell, which performs the coherence function of the glutathioneuretase enzyme that provides its synthesis. It is also a metabolite that is essential for normal functioning of catalase enzyme. Catalase, which takes part in many processes, is one of the components of the cell defense system. NADPH is also used in the synthesis of steroids which play an important role in its completeness and functional activity, which is one of the components of the membrane. One of the important places to use it is the synthesis of fatty acids. The localization of the DMDH enzyme in plant tissues coincides with almost the localization of Q6PDH enzyme. Given that the main product of both enzymes is NADPH, it can be assumed that it will

participate in functions which are specific to the Q6PDH enzyme associated with this metabolite.

MATERIAL AND METHODS

In order to study the effects of salt stress on the growth dynamics of sprouts, germinated seeds intended as control variants were cultured in distilled water and experimental varieties were cultivated at 25 ° C for 7 days in appropriate saline solutions. The ages and sizes of the root and hip systems of the sprouts were determined on days 3, 5, and 7 of the cultivation.

One of the key points in determining the activity of the enzymes is the correct selection of the extraction environment. For this purpose, 0.1 M tris-HCl buffer solution containing 1% polyvinylpyrrolidone (24 kDa mass) was used to neutralize the 0.01 M ß-mercaptoethanol and the phenolic compounds as a reducing agent in our experiments. Based on literary data, the pH value for extracting Q6PDH enzyme was 8.0 and the buffer solutions of pH 7.0 for extraction of DMDH enzyme were taken. In the preparation of homogenate, 1 ml of biological object was extracted with 2 ml of extraction solution and crushed in a cold environment using an ice bath. In both cases, the homogeneous homogeneity obtained from the double caprous tissue was collected at centrifuge at a speed of 5,000 rpm for 10 minutes and was used to determine the activity of the supernatant. The enzyme preparations made in this way had a stable activity in cold weather for several hours and did not have any difficulties in the measurement.

The activity of both enzymes was determined by spectrophotometric method, at 340 nm wavelength, based on NADP reducing rate. The weight of ΔE103340 / min / q / min / g was taken as an enzyme unit.

RESULTS AND DISSCUSSION

Influence of the cytoplasmic G6PDH and DMDH enzymes on the activity dynamics and the ratio of their activity to beans of bean seeds of NaCl, Na2SO4, Na2HCO3 and Na2CO3 salt solutions are shown in Table 1 below.

When the results presented in the table are compared to those obtained with wheat and barley crops, it is easy to conclude that the class of doubles and the level of activity of G6PDH and DMDH enzymes in the root system tissues of bean herbaceous plants, which are relatively sensitive to salt stress and stress, are significantly lower than the root system tissues of wheat and barley seedlings. Perhaps, there is a certain link between the activity levels of these enzymes and their durability. However, the imposition of such a provision requires a large number of additional experiments to clarify this problem.

Table 1

Due to the development of wheat seeds of Na-isocated salt solutions

Influence of Q6PDH and DMDH enzymes on activity dynamics

Variants	Q6PDH activity			DMDH activity			Q6PDH/ DMDH		
	3 day	5 day	7 day	3 day	5 day	7 day	3 day	5 day	7 day
Kontrol	62	83	101	31	38	43	2.00	2.18	2.35
NaCl									
25 mM	75	99	137	36	47	59	2.08	2.11	2.32
50 mM	81	108	149	42	55	61	1.93	1.96	2.44
100 mM	85	93	98	47	56	68	1.81	1.69	1.44
Na_2SO_4									
25 mM	83	107	148	39	51	67	2.13	2.10	2.21
50 mM	94	115	123	48	57	61	1.96	2.02	2.02
100 mM	70	73	61	51	61	53	1.37	1.20	1.15
$NaHCO_3$									
25 mM	70	83	98	47	63	72	1.49	1.32	1.36
50 mM	75	88	85	59	71	75	1.27	1.24	1.13
100 mM	69	60	59	61	60	55	1.13	1.00	1.07
Na_2CO_3									
25 mM	73	85	96	51	65	68	1.43	1.31	1.41
50 mM	81	77	65	63	76	50	1.29	1.01	1.30
100 mM	–	–	–	–	–	–	–	–	–

The activity of the cytoplasmic Q6PDH enzymes in the distilled water (control variant) bean seeds is significantly increased due to the extension of the incubation period, unlike the cytoplasmic Q6PDH enzymes of wheat and barley seeds. Thus, in the root system of the 5-day seedlings, activity was 33.9%, and 62.9% higher in the root system of 7-day seeds compared with the 3-day spruce roots.

There is also an increase in DMDH activity in the development of the root system, but compared to the Q6PDH enzyme, this increase appears to be relatively weak in its manifestation. Thus, the activity of DMDH enzymes increases by 22.6% in the 5-day period and 38.7% in the 7-day sprout, compared to 3-day seedlings. In spite of the high figures in terms of interest, in fact, this increase is only 7 Δ10 for 5-day seedlings and 3340 / min for the 7-day seedlings and 12 Δ10 • 3340 / min / h. Bean seeds are different from wheat and barley seeds in this regard. As you have already seen, the activity of the enzyme is intensely induced by the development of the root system.

Such a change in activity is also reflected in Q6PDH / DMDH. For 3-day seedlings, this figure is characterized by 2.00, 5-day seedlings 2.18, and 7.35 seeds with 2.35. That is, Q6PDH enzymes were more active than the DMDH enzyme at all times observed in the development of the root system of the seedlings, and apparently, Q6PDH is more important in shaping the NADPH pulse in the root system tissues of the bean herb.

The effect of NaCl salt solutions on the Q6PDH enzyme activity dynamics is directly related to their density and duration. During the short-term effect (3-day seedlings) there is a positive relationship between the concentration of the salt and the induction of the activity of the enzyme, ie the degree of induction of ferment activity increases as the solids in the solution increase. In 5-day sprouting, this pattern is essentially

true at 25 and 50 mM, and the induction rate of the enzyme at 100 mM has already started weakening and its activity is relatively low in comparison with the density of 25 and 50 mM and relatively high in the control variance. Increasing the incubation period up to 7 days leads to an increase in induction in thicknesses at 25 and 50 mM and a significant reduction in the 100 mM concentration, and, eventually, the activity decreases from the activity level of the control peripheral.

NaCl salt solutions lead to substantial stimulation of the activity of DMDH enzyme. As the duration of the salt increases and the duration of its expiration, the induced effect increases. Compared to the Q6PDH enzyme, NaCl salt solutions lead to a relatively more intense induction of DMDH enzyme activity and considerably increase the Q6PDH / DMDH activity. That is, the price of Q6PDH / DMDH is lower as the NaCl salt increases in the solids. This, in turn, indicates an increase in the role of DMDH enzyme in this process in the stress conditions generated by NaCl salt.

In analogous concentrations, Na2SO4 salt solutions have a more potent stress condition than NaCl saline solutions, or beans have a more adverse effect on the growth of the seeds, as well as the activity of the enzymes. The effect obtained with NaCl salt solutions is then observed in a lower density of the salt. This leads to the fact that the activity of the Q6PDH enzyme is intensely induced at all levels of incubation at relatively low concentrations (25 and 50 mM), while the effect of the upper layer (100 mM) is weakened (at 3 md in mM) induced effects (5 and 7 day seeds) are replaced by ingestion effect. As can be seen from the figures presented in the table, the activity level of the enzyme in the 3-day seedlings of 100 mM is higher than the control variant, and the experimental variant is at a low level. However, the prolongation of the incubation curve also results in lower levels of activity than control.

Na2SO4 salt solutions also strongly inhibit the activity of DMDH fertilizer relative to NaCl saline solutions. In this case, the weakening of the induction effect is only observed in the high solids of the salt and at the end of the incubation. Indeed, in the experimental version, the activity remains significantly higher than that of the control period of the relevant period. The high stress factor generated by the Na2SO4 salt solutions is negative to the activity of the Q6PDH enzyme, and the positive effect on the activity of DMDH enzyme is lower than that of the Q6PDH / DMDH indicator in NaCl saline solutions. That is, the contribution of DMDH enzyme to anesthetization to salt stress is rising, and it begins to play an important role in this process.

NaHCO3 and Na2CO3 salt solutions, which have a more adverse effect on the development of the bean seedlings, have a significantly different effect on the activity dynamics of the root system's Q6PDH and DMDH enzymes than the NaCl and Na2SO4 salts. Their solutions were only caused by the inhibition of Q6PDH enzyme activity in low density and short-term effects. Increasing the thickness and duration of the injection resulted in the weakening of the induction effect and its replacement with ingibiration effect. For example, if the NaHCO3 salt solution absorbs the Q6PDH activity of 25 mM in the 3-day seedlings in 12.9%, 50 mM density at 21.0%, 100 mM 11.3%, and the dose of 25 mM in 7 days of germination the positive rise in the dynamics of growth, gradually slowed down, gradually slowed from 101 Δ10 • 3340 / min to 98 Δ10 • 3340 / min, and due to the increased density, this process was sharply strengthened with a control variant of the same period of 50 mM compared to 15.8% in comparison with 41.6% in 100 mM density. Similar results were also obtained for Na2CO3 salt solutions. His ingibirating effect on this process was observed in lower concentrations compared to NaHCO3 salt solutions.

Unlike the Q6PDH enzyme, the activity of the DMDH enzymes of the root system of the bean seedlings in the NaHCO3 and Na2CO3 salt solutions has been substantially induced in all segments, irrespective of the duration of the incubation (ie all periods of incubation). In other words, in the experimental variants, the activity of the enzyme was always higher than control. For example, the root system of incubated seedlings in the 25 mM NaHCO3 salt solution in 3-day seedlings has increased 51.6% in the tissues of activity. The similar increase was 90.3% for 50 mM and 96.8% for 100 mM. In subsequent incubation periods, the relatively low concentrations of the salt solution (25 and 50 mM) further enhanced the activity of the enzyme, while the induction effect in the high layer (100 mM) was relatively weakened. The similar effect of Na2CO3 salt solutions to this process was observed in lower concentrations than NaHCO3 salt solutions.

This difference in the NaHCO3 salt solutions in the activity doses of Q6PDH and DMDH has also been reflected in the Q6PDH / DMDH value. This indicator which changes in the range 2.00-2.35 due to the development of control variant variants, decreased to 1.00-1.13, due to the stress condition deterioration.

The analysis of received results suggests that Q6PDH plays a major role in NADPH synthesis in relatively poor stress caused by Na-isocationic saline solutions in the root system tissues of the bean seeds, while DMDH enzyme in relatively acute stress conditions.

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