



Study of enzymes that play a role in the defense mechanism against abiotic factors in dicotyledonous plants

Asadova Basti Goshun

Azerbaijan State Pedagogical University, Baku, Azerbaijan

Abstract

Abiotic stress is the major cause of crop loss worldwide, reducing average yield of plants by more than 50%. Plants are subjected to many abiotic stresses like drought, heat, low temperature and salinity stress. Plants have adopted to live in environments where they are often exposed to different stress factors in combination. They have developed specific mechanisms that allow them to sense particular environmental changes and act in response to complex stress conditions, reducing damage while conserving important resources for growth and development. Plant triggers a specific and unique stress response when subjected to different environmental stresses. The response to change in environment can be quick, depending on type of stress and can involve either adaptation mechanisms, or avoidance mechanism.

Keywords: abiotic stress, plant, plant enzymes, bean, pea, seed

Introduction

Abiotic stresses, such as temperature extremes, drought, salinity, and heavy metals are major factors limiting crop productivity and sustainability worldwide. Abiotic stresses disturb plant growth and yield formation. Several chemical compounds, known as plant growth regulators (PGRs), modulate plant responses to biotic and abiotic stresses at the cellular, tissue, and organ levels. Thiourea (TU) is an important synthetic PGR containing nitrogen (36%) and sulfur (42%) that has gained wide attention for its role in plant stress tolerance. Tolerance against abiotic stresses is a complex phenomenon involving an array of mechanisms, and TU may modulate several of these. An understanding of TU-induced tolerance mechanisms may help improve crop yield under stress conditions. However, the potential mechanisms involved in TU-induced plant stress tolerance are still elusive. In this review, we discuss the essential role of TU-induced tolerance in improving performance of plants growing under abiotic stresses and potential mechanisms underlying TU-induced stress tolerance. We also highlight exploitation of new avenues critical in TU-induced stress tolerance.

The negative effects of salinity stress are manifested in all areas of plant metabolism as a whole. It affects the metabolism of many intracellular substances, including nucleic acids, proteins, carbohydrates and amino acids. In the course of evolution, plants have created various mechanisms to protect against the hyperosmotic situation caused by salinity and to disturb the balance of mineral metabolism, and to continue the struggle for survival. Most of these protective reactions are associated with the expression of genes and are accompanied by certain changes in the metabolic process, which also help the plants to restore the disturbed state of hemostasis.

Material, Methods and Results

The effect of NaCl, Na₂SO₄, NaHCO₃ and Na₂CO₃ salt solutions on the activity dynamics of cytoplasmic G6PDH and DMDH enzymes of bean and pea seed roots is reflected below.

Comparing the results with similar results obtained with wheat and barley, it is easy to conclude that the activity of both G6PDH and DMDH enzymes in the root system tissues of bean sprouts belonging to the dicotyledonous class and considered relatively sensitive to salt stress is wheat. and significantly lower than the root system tissues of barley seedlings. It is possible that there is a link between the activity levels of these enzymes and their salt resistance. However, the introduction of such a provision requires large-scale additional experiments to clarify this problem.

Table 1

Variants	G6PDH activite			DMDH activite			G6PDH/ DMDH		
	3day	5 day	7 day	3 day	5 day	7 day	3 day	5 day	7 day
Control	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 ₂ SO 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 ₂ CO 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 cytoplasmic Q6PDH enzyme in the roots of bean seeds grown in distilled water (control variant) is significantly increased due to the prolongation of the incubation period, unlike the cytoplasmic Q6PDH enzyme in the roots of wheat and barley seeds. Thus, compared to 3-day-old seedlings, the activity of 5-day-old seedlings in the root system was 33.9%, and in the root system of 7-day-old seedlings was 62.9% higher.

There is also an increase in DMDH activity associated with the development of the root system, but this increase is relatively weak compared to the enzyme Q6PDH. Thus, compared to 3-day-old seedlings, the activity of DMDH enzyme in 5-day-old seedlings increases by 22.6%, and in 7-day-old seedlings by 38.7%. Although the numbers are high in percentage terms, in fact, this increase is only $7 \Delta 10 \cdot 3340 / \text{min} / \text{g}$ for 5-day seedlings and $12 \Delta 10 \cdot 3340 / \text{min} / \text{g}$ for 7-day seedlings. In this respect, bean seed seedlings differ from wheat and barley seed seedlings. As already mentioned, they induce the activity of the enzyme more intensively due to the development of the root system.

This change in activity is also reflected in the Q6PDH / DMDH ratio. This figure is 2.00 for 3-day-old seedlings, 2.18 for 5-day-old seedlings, and 2.35 for 7-day-old seedlings. In other words, the Q6PDH enzyme has been more active than the DMDH enzyme in all periods observed in the development of the root system of seedlings, and it seems that Q6PDH is more important in the formation of NADPH money in the root system tissues of bean seedlings.

Since Na₂SO₄ salt solutions at similar concentrations create stronger stress conditions than NaCl salt solutions, they have a more negative effect on both the development of bean seed seedlings and the dynamics of enzyme activity. The effect obtained with NaCl salt solutions is observed in this case at a lower concentration of salt. This leads to the fact that at relatively low concentrations (25 and 50 mM) the activity of the enzyme Q6PDH is sharply induced during all periods of incubation, and at high concentrations (100 mM) this effect is either weakened (in mM in 3-day seedlings) or the induction effect (in 5- and 7-day seedlings) is replaced by the inhibition effect. As can be seen from the figures presented in the table, the level of enzyme activity in 3-day-old seedlings at a concentration of 100 mM is higher than in the control variant and lower than in the experimental variant. However, prolonging the incubation period causes activity to fall below control levels.

Unlike the Q6PDH enzyme, the activity of the DMDH enzyme in the root system of bean sprouts under the influence of NaHCO₃ and Na₂CO₃ salt solutions was significantly induced at almost all concentrations, regardless of the duration of exposure (ie during all incubation periods). That is, in the experimental variants, the level of enzyme activity was always higher than in the control. For example, in 3-day-old seedlings, activity in the root system tissues of seedlings incubated in 25 mM NaHCO₃ salt solution increased by 51.6% compared to control. A similar increase was 90.3% for 50 mM and 96.8% for 100 mM. In subsequent incubation periods, relatively low concentrations of the saline solution (25 and 50 mM) further enhanced the enzyme activity, while at higher concentrations (100 mM) the induction effect was relatively weak. A similar effect dynamics of Na₂CO₃ salt solutions was observed at lower concentrations than NaHCO₃ salt solutions.

Such a different effect of NaHCO₃ salt solutions on the dynamics of activity of Q6PDH and DMDH enzymes is reflected in the value of Q6PDH / DMDH. This indicator, which varied in the range of 2.00-2.35 due to the development of seedlings of the control variant, decreased to 1.00-1.13 due to the aggravation of stress.

Analysis of the obtained results suggests that Q6PDH plays a key role in the synthesis of NADPH under relatively low stress conditions caused by Na-isokationic saline solutions in the root system tissues of bean seedlings, and DMDH enzyme under relatively severe stress.

Recent research suggests that both enzymes play an important role in the ability of plants to respond to and adapt to the extreme conditions of the environment in which they live. The participation of both enzymes in this process is based on the synthesis of the NADPH molecule due to their catalytic effect on their substrates. NADPH is shown to be involved in the neutralization of many biochemical and, in this connection, a number of physiological factors, including the negative effects of extreme factors.

Table 2 below shows the results of experimental studies on the effect of Na-isoquated salt solutions on the dynamics of activity of cytoplasmic G6PDH and DMDH enzymes and their activity ratio in the root system of pea seedlings.

It is easy to compare the figures presented in the table with the similar figures obtained from the study of the root system of the seedlings already considered.

It can be seen that the enzymes Q6PDH and DMDH occupy an intermediate position between wheat sprouts and bean sprouts in terms of the level of activity in the tissues of the root system of pea seedlings, one of the representatives of other dicotyledonous plants that are more resistant to salt than beans.

Table 2

Variants	Q6PDH activite			DMDH activite			Q6PDH/ DMDH		
	3 day	5 day	7 day	3 day	5 day	7 day	3 day	5 day	7 day
control	78	99	113	53	59	67	1.47	1.68	1.69
NaCl									
25 mM	93	118	127	61	66	70	1.52	1.79	1.81
50 mM	107	127	137	65	71	75	1.65	1.79	1.83
100 mM	116	121	110	71	76	83	1.63	1.59	1.33
Na ₂ SO ₄									
25 mM	99	131	145	66	69	75	1.50	1.90	1.93
50 mM	113	135	122	70	73	87	1.61	1.85	1.40
100 mM	121	116	101	77	81	96	1.57	1.43	1.05
NaHCO ₃									
25 mM	86	109	112	77	98	109	1.12	1.11	1.03
50 mM	91	110	93	91	86	99	1.00	1.28	0.94
100 mM	103	91	78	99	85	68	1.04	1.07	1.15
Na ₂ CO ₃									
25 mM	89	112	105	81	96	91	1.10	1.17	1.15
50 mM	93	85	89	85	81	75	1.09	1.05	1.19
100 mM	--	--	--	--	--	--	--	--	--

As can be seen from the figures presented in the table, as in the case of bean seedlings, the activity of the Q6PDH enzyme gradually increases with the development of the root system of pea seedlings in the control variant and reaches its maximum in the root system of 7-day seedlings. During this period, the activity of the enzyme increases by 36.6% compared to 3-day seedlings. Due to the development of the root system, there is an increase in the activity of the enzyme DMDH, although weaker than the enzyme Q6PDH. In this case, a similar increase in 7-day seedlings compared to 3-day seedlings is 14.8%.

Due to the development of the root system of seedlings, certain changes occur in the Q6PDH / DMDH indicator. In contrast to the results of experiments with wheat and barley, the value of this indicator in the root system of pea seedlings does not decrease, but rather increases. If for the root system of 3-day-old seedlings it was 1.47, in the root system of 7-day-old seedlings it gradually increases to 1.69. In other words, the role of the enzyme Q6PDH in the formation of NADPH potential due to the development of the root system of pea seedlings not only decreases, but rather increases.

Thus, the following main conclusions characterizing the root system of pea seedlings can be drawn: first, the activity of Q6PDH enzyme in these tissues is significantly higher than the activity of DMDH enzyme, and secondly, the involvement of both enzymes in NADPH enrichment of root system cells under salt stress. however, the role of the Q6PDH enzyme increases mainly under relatively low stress conditions and the DMDH enzyme under severe stress conditions under Na-isolated salt solutions.

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