

## BIOCHEMICAL MECHANISMS THAT ARE INVOLVED IN THE PROCESS OF ADAPTATION OF PLANTS TO ENVIRONMENTAL CONTAMINANTS

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### Abstract

Two aspects of biochemical shifts in plants during adaptation to contamination are speculated: amino acid exchange in wild and cultural cereals as phytoextractors of heavy metals from soil and surface lipids of Coniferous growing on industrially contaminated areas. Total quantity of free amino acids in primary leaves of cereals increased under influence of heavy metal ions in some species in 10 times. We proposed the value that may describe not only quantity of free amino acids but the state of protein exchange – *index of unincluding* – ratio between quantity of certain amino acid in free and bound state. Phytoextraction of heavy metals is followed by significant rebuilding of protein exchange in tissues of cereals that is shown and discussed. Surface lipids (SL) is a protective layer of lipid molecules (epicuticular lipids, surface waxes). We found that composition of SL changes with age and under industrial contaminants. Two elongation-decarboxylation (ED) systems in surface tissues of plants work independently: EDI – lead to hydrocarbons, free fatty acids with odd-carbon chain (free components of SL) and EDII – lead to esters (bound components of SL). The conclusion was made: EDI-pathway is more sensitive than EDII one but they both make impact to adaptation mechanisms of plants.

### Introduction

This paper is devoted to two independent biochemical investigations: one (A) is connected with amino acid exchange in cereals under influence of soil contamination and another (B) with surface lipids of plants and their formation under influence of some environmental factors.

A. It is well known that some plant species can adapt to pollution by developing protection mechanisms such as increasing of free amino acid pool in their tissues and root exudates that plays an important role in sequestration of the pollutants in the roots, transportation and immobilization in the vacuoles or in the cell walls [1, 2]. Here we focus our attention on amino acid exchange of plants, especially on some mechanisms of free amino acids formation and their possible functional role in plant tissues.

B. Surface lipids (SL) – a layer of lipid molecules (epicuticular lipids, surface waxes) that protect plants from UV-irradiation, hydrophobic pollutants and diseases. Our previous investigations [9 - 11] showed that SL of leaves and seeds of plants had esters, hydrocarbons, free fatty acids, oxo-substances (some species) etc. Content of SL is varying in species, changes with age and under influence of pollution. Different types of plants choose certain biosynthetic strategy to build a lipid barrier between living cells and environment.

### Materials and methods

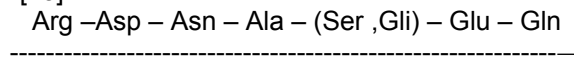
A. Amino acid composition of proteins and free pool of seeds, seedlings, adult plants of different genetic forms of *Zea mays L.*, and wild cereals *Festuca rubra L.*, *Lolium perenne L.* was studied and described in [3 – 8].

B. Surface lipids (SL) were obtained from leaves of Coniferous, liana *Hoya carnosa L.f*, tropical plants, grain of different types of maize and sorghum. Procedures of extraction and investigation of surface lipids are described in [9 - 11].

### Results and discussion

A. There exists certain disparity between amino acid content of plant proteins and free pool in tissues that confirms the multifunctional role of these substances in the cell.

In the rape grain of maize of different genetic forms you may find total quantity of free amino acids: from 0,5 to 12 g per 100 g of protein or from 70 to 15000 mg% on dry weight. The disparity in content between free and bound amino acids is more during the processes of germination and maturing [8]. It was found certain reverse correlation between quality of stored proteins and size of amino acid pool, that was an explanation of mechanism of biosynthesis of stored proteins in high lysine mutants [12]. The osmotic and regulatory role of amino acids in free state was the mane position of these works. The surplus of amino acid pool in grain corresponds to ability to be attacked by fungi and insects and promotes to worse storage of seeds [8]. Calculations of energetic provision of tissues made possible to put amino acids that are accumulating in free state in the row [13]:



As  $[ \text{ATP} ] > [ \text{ADP} ] + [ \text{AMP} ]$ , i.e. Atkinson's charge of a cell is large, glutamate family of amino acid is accumulated and if the cell is energetically exhausted, family of aspartate is accumulated; and the same considerations of the middle states.

Free amino acids in leaves correlate with maturation of reproductive parts of plants as they are the main nitrogen containing substances required by storage proteins, reflect the process of senescence of leaves, are regulators of a lot of biochemical processes.

The surplus of free amino acid pool may be the result of reducing or of intensification of protein biosynthesis, some disturbances in transport of amino acids, and (or) in all processes where amino acids take part. It is a nonspecific (in some respect) reply of a plant cell to any influence, positive or negative. The most relevant value that may describe not only quantity of free amino acids but the state of protein exchange in a whole may be an *index of unincloding* (IU)– ratio between quantity of certain amino acid in free and bound state in %. Using this value it is possible to demonstrate abundance of some amino acids in plant tissues. The most frequently the abundance, or electoral accumulation, of Asn + Asp, Gln + Glu, Pro take place in plant tissues that is discussed earlier [4 –7]. During grow of seeds of wild grasses on contaminated with heavy metals soils in conditions, described in [1], total quantity of amino acids in primary leaves increased and electoral accumulation of Phe, Tyr and Ser was found, that depended from a specie (fig.1).

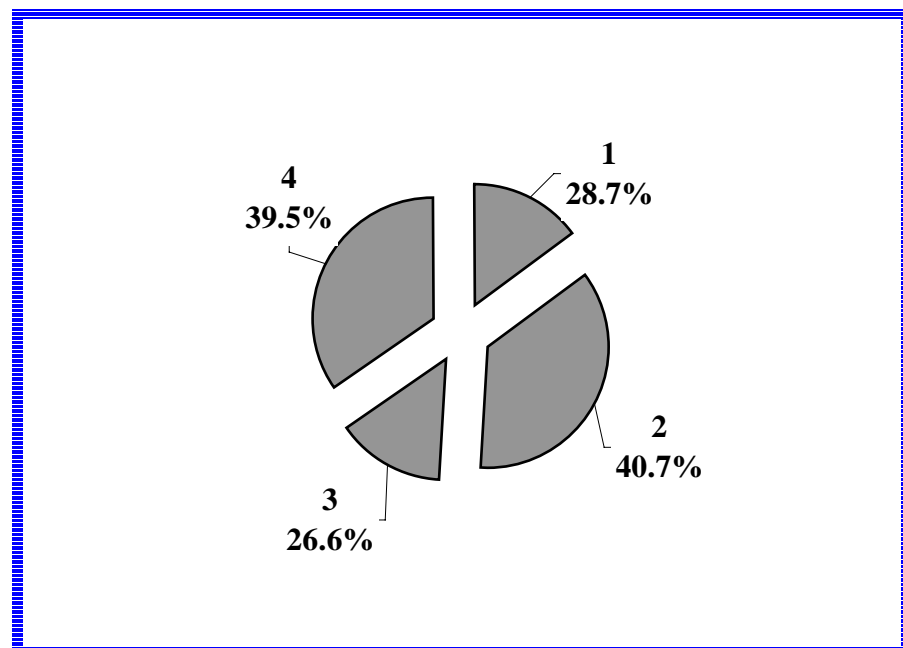


Fig.1. Indexes of unincloding of free Tyr (1, 3) and Phe (2, 4) in primary leaves of *Lolium perenne* L. in experiments with zinc salts without (1, 2) and with growth regulator FeSuc (3, 4)

It is known that aromatic amino acids are synthesized in plants from shikimic acid, which is formed from glucose:

D-Glu → → → 5-Phosphoshikimic acid → Prephenic acid → Phenylpyruvate → **Phe**

Synthesis of Tyr may take place as on main rout:

Prephenic acid → p-Hydroxyphenilpyruvate → **Tyr**

or on additional rout:

Prephenic acid → Pretyrosine → **Tyr**

These two amino acids are precursors of flavonoids that play an important role in the processes of adaptation of wild cereals to environmental factors and their biosynthesis is more active in these species than in cultural ones. As there are some experiments, where quantities of Tyr are more than Phe, that is very rare for plant tissue, we consider that both routs of synthesis of Tyr are active in stress conditions.

Ser is synthesized in plant tissues from the products of Calvin cycle by three routs. We guess that the most active is the way through phosphoglycerate .

B. Investigation of SL of plants shows different content of classes and individual components in a certain class of lipid molecules. For example, hydrocarbons and fatty acid content of SL of Coniferous differ very much from that of liana *Hoya carnosa* and from SL of grain of maize and sorghum. But the most differences among species found by us is content of oxo-substances, or aldehydes and ketones. Leaves of Coniferous don't contain them at all, while SL from leaves of tropical trees contain a lot of them; SL from grain of maize have some little quantities while sorghum have them as a main class of SL. DTA is a sensitive method that describes molecular-dynamic characteristics of molecules and may be used for investigations of SL [9]. Two endoeffects were found in the SL of investigated Coniferous. First low temperature endothermic effect was found only for control plants and has no minimum. The second endothermic effect had a sharp minimum and was found for all species both from control and contaminated area. It was shown that the biosynthesis of SL is an active process during whole living period of a plant and is especially intensive in surface tissues of young leaves. Endothermic effect I is low temperatural, has no minimum and according to literature data means melting of amorphous phase of SL. Absence of it in SL of plants grown on contaminated territories may be explained by reducing of biosynthesis of hydrophobic components (hydrocarbons, esters) of SL under influence of chemical volatile substances or soil contaminants that are present in industrial regions. Second endothermic effect characterizes transition solid – liquid state of crystalline phase of SL. It has sharp minimum that is common for crystalline compounds and is relevant to more polar components of SD, free fatty acids for example. Under influence of toxicants we found changing of parameters of this effect: it has wider borders (it means that heterogeneity of polar substances is more than in SL of control ones) and the temperature of minimum is higher (it means increasing of content of polar compounds which are able to associate). These data are confirmed with data of UR-spectroscopy [10], where we had observed the increase of carbonyl absorbtion in SL of plants from industrial regions. As mentioned above two elongation-decarboxylation systems ( EDI lead to hydrocarbons, alcohols, free fatty acids with odd-carbon chain and EDII lead to esters) both synthesize hydrophobic and polar components, they both make impact to adaptation mechanisms of plants. SL from seeds of sorghum had not I endothermic effect while SL from maize seed had both of them. As these species differs on content of oxo-substances, we think that polarity of components and the length of hydrocarbon chain are two factors that determine the DTA characteristics of SL of plants.

## Conclusions

Change of free amino acid content and quantity is a widespread mechanism of adaptation of plants to environmental factors. Being studied is a good tool for phytoremediation and phytochemistry. Spectral and DTA data confirms the fact that surface lipids of plants take part in the process of adaptation to environmental factors. The direction of rebuilding of surface layer under contamination is biosynthesis of more polar surface lipid molecules.

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