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METABOLOMICS AS A MODERN APPROACH FOR THE INVESTIGATION OF POTATO PLANT ADAPTATION TO BIOTIC AND ABIOTIC STRESS FACTORS

(review)

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Abstract

The progress in genomic and proteomic investigations has greatly expanded the range of subjects aimed in discovering of mechanisms involved in the regulation of plant growth and development under changing of environmental conditions. Another systemic biology approach, which is known as metabolomics, has almost the same significance. It focuses on the study of dynamics of low molecular compounds which results from the complex metabolic processes in the cell. The intensity of these processes is under the influence of both biotic and abiotic stress factors. Studies on metabolic analysis are carried out not only with model objects, but also with cultivated plants, including potatoes, listed among top 10 of the most valuable crops. This review aims to summarize the available literature data on systemic biochemical rearrangements detected with metabolic approach in potato under the action of pathogenic viruses and microorganisms, insects, as well as under the influence of abiotic stressors on potato plants. Recent data indicates that metabolic analysis allows characterization of the development and progression of viral and bacterial diseases, as well as testing resistance to the infections in various potato species and varieties (H. Hamzehzarghani et al., 2016; T. Stare et al., 2015; H. Tai et al., 2014; S. Tomita et al., 2017). Significant changes in a number of secondary metabolites are shown. The metabolic approach has sufficient sensitivity to detect also alterations under environmental stress. In the review, it was considered that the results of metabolic rearrangements of the potato cell are directly linked to dehydrogenation, including osmotic and temperature stressors. The changes in the content of amino acids and sugars are of particular importance. However, a number of additional studies are required for evaluation of shifts in potatoes metabolism which are triggered under the combined stress factors action, for example, desiccation and hyperthermia (V. Arbona et al., 2013; M. Drapal et al., 2017; R.D. Hancock et al., 2014). An absolute majority of the metabolic data was obtained with various vegetative organs of potato plants. Unfortunately, metabolic profiles of generative organs have not been studied yet. There is no information on the metabolic profiling of pollen formation, including CMS-forms of potatoes. This indicates the importance of this direction in the investigation of potato metabolome. Further standardization of the metabolic analysis and methods of result processing will make it possible to use the metabolomics not only as an important component of fundamental research, but in time, as a basis for monitoring of collection samples and newly created varieties and hybrids of potatoes. Analysis of modern data indicates their perspective for phenotyping of different potato genotypes, as well as for identifying forms that are resistant to various types of unfavorable conditions.

Keywords: metabolomics, *Solanum* spp., potato, biotic stress, pathogens, viral infection, fungal infection, pests, abiotic stress

Plants synthesize plenty compounds of various chemical structures. These compounds are generally grouped as those of the primary metabolism which ensure the existence of any living being, and of the secondary metabolism which are characteristic of certain (sometimes very limited) groups of organisms.

The number of plant metabolites exceeds 100,000, and only a small part of these substances are currently identified [1]. Substantially intensified research on their diversity became possible due the novel methods that allow characterization of all metabolites of a biological system, i.e. an organism, an organ, a tissue, etc. [2, 3]. Recently, gas chromatography—mass spectrometry (GC-MS), high-performance liquid chromatography—mass-spectrometry (HPLC-MS), ultra-performance liquid chromatography tandem mass-spectrometry (UPLC-MS), capillary electrophoresis—mass spectrometry (CE-MS) and nuclear magnetic resonance (NMR) spectroscopy are used to separate and identify metabolites [4, 5]. Advanced technologies and uniform extraction and detection methods, and available databases for the identification of various compounds, as well as multivariate statistics methods provide excellent opportunities for comparing metabolite profiles, which is the basis of metabolomics.

Metabolite accumulation patterns determine significance of crops in the production of food and raw materials for the pharmaceutical, chemical and other industries [5]. Thence, metabolite data are not only of theoretical, but also of practical significance, and crop plants, in particular potato (*Solanum tuberosum* L.) cultivars, become models. Global annual yield of potatos reaches 300 million tons, which determines the role of potato cultivars for food security [6]. One of the first metabolomic studies on potato plants performed in 2000 using GC-MS [7] was focused on the analysis of carbohydrate metabolism compounds. The authors found differences between the tubers of plants grown in vitro and in the field conditions, as well as between wild type plants and transgenic lines [7]. Further metabolite profiling revealed differences between organs (for example, leaves and tubers responsible for the primary synthesis of carbohydrates and their deposition) and proved that metabolomics is a sensitive tool to evaluate genetic modifications and to control breeding [8].

The present review focuses on an analysis of current metabolomic approach to study mechanisms of adaptation to biotic and abiotic stressors of potato plants and to identify their resistance.

Effect of biotic agents. Plants that during growth interact with a huge number of organisms, including pathogens, have developed resistance mechanisms in the course of evolution. Omix technologies can trace manifestation of such mechanisms at different levels, i.e. from genome (differentiated gene expression, including the family of *PR* genes) through the proteome (production of protective proteins and enzymes for the synthesis of low molecular weight compounds) to metabolome (change in alkaloids, substances of phenolic nature, etc.) [1, 9]. In cultivated plants, secondary metabolites which often are the compounds involved in protection against infection, can significantly reduce the nutritional value of products or even are poisonous [10, 11].

Viral infections. Potato is infected by at least 40 viruses (http://www.kartofel.org/bolezn/virus/virus.htm). A large-scale studies revealed seven main types of potato resistance to viruses [12, 13], including resistance to infection (field resistance), resistance to virus accumulation, restriction of virus transport, resistance of mature plants, tolerance, resistance to vectors of viruses and hypersensitivity. Many plant systems at different levels, from cellular to organismic, are involved in each type of resistance. Recent data indicate that the virus attack initiates changes in the activity of a number of metabolic pathways, including carbohydrate metabolism and the synthesis of amino acids [14-16]. Consequently, metabolomics can be a tool to study mechanisms of resistance to viral infection in potato species and varieties.

Potato virus Y (PVY, *Potyviridae*) is among the most economically significant because of yield losses up to 80 % in susceptible varieties. PVY was first

described more than 80 years ago as an agent that causes degenerative processes in potatoes [17, 18]. The first data on modifications of the primary metabolism when exposed to the aggressive PVY^{NTN} isolate was obtained for potato varieties Desiree [19]. It was found that these processes are connected with the reproduction of the virus in the tissues. The authors studied predominantly carbohydrate metabolism and showed a correlation between a decrease in carbohydrate content and a change in the expression of genes encoding proteins of the photosynthetic apparatus and photoassimilation enzymes. In transgenic plants of the Desiree cultivar with overexpression of salicylate hydroxylase *NahG* gene from *Pseudomonas putida*, inhibition of photosynthesis was more pronounced and the amount of salicylic acid, a biotic stress hormone, lowered [19].

GC-MS metabolomic profiling identified 168 metabolites in dynamics during PVY infection in potato leaves and revealed differences between clusters associated with synthesis of amino acids and secondary metabolites, synthesis and degradation of cell wall substances [9]. Principal component analysis (PCA) showed clustering corresponding to the time of virus infection. The plant response to infection varied notably on day 3 and was more uniform on days 1 and 6. Analysis of variance (ANOVA) showed statistically significant changes ($p < 0.01$) for 83 metabolites, including 32 those identified, which depend on the stage of infection and PVY strain (PVY^N or PVY^{NTN}). PVY^N infection is asymptomatic, and PVY^{NTN} causes a mosaic, chlorotic and necrotic lesions of leaves, as well as tubers. In PVY^{NTN} infection, γ-aminobutyric acid (GABA), α-ketoglutarate, glycerate, maleate, maltose, phenylalanine, pyruvate, succinate, sucrose, and valine were varying metabolites. The amount of sucrose, glycerate, succinate and threonate decreased statistically significantly at the beginning of the disease in infected leaves as compared to the control ones. In PVY^N infection, the sucrose content also decreased significantly, despite the fact that this isolate is less aggressive. A similar dynamics was observed for most other metabolites involved in amino acid metabolism, the Krebs cycle, GABA shunting, and neutralization of reactive oxygen species (ROS) and phenylpropanoids. Valine and compounds associated with the metabolism of phenylpropanoids were found in infected leaves in larger amounts on day 6 when intensive reproduction of the virus occurred. PVY^N, in contrast to PVY^{NTN}, causes earlier accumulation of ROS neutralizing compounds in leaves, which is probably due to less damage caused by the less aggressive PVY^N strain [9].

The metabolomic changes are in line with the data of transcriptome analysis. The changes in carbohydrate metabolism, GABA shunting, phenylpropanoid and antioxidant metabolism depend on the period of the infection and the viral strain [9]. Accordingly, metabolic analysis is suitable for detection of viral infection and plant responses.

Fungal infections. Late blight. Phytophthora infestans is among the most harmful fungal pathogens of potatoes. This infection can lead to complete loss of potato crop [20, 21]. Fungicides which are used against late blight are quite expensive and negatively affect the environment. In addition, their regular use stimulates pathogen resistance. Growing varieties resistant to the pathogen is an alternative [22]. Potato plant resistance to late blight is controlled by many interacting genes and quantitative trait loci (QTL) some of which determine biochemical traits [23, 24]. Metabolomics can be a tool for describing biochemical changes and identifying markers of resistance in fungal infections [25].

Analysis of metabolite profiles of the aboveground parts of potato plants contrasting in susceptibility to phytophtore under controlled conditions show that phenylpropanoids, especially hydroxycinnamic acid amides, flavonoids, alkaloids and fatty acids the synthesis of which is more strongly induced after infection of resistant lines are primarily associated with resistance. During pathogenesis, ex-

pression of genes involved in the biosynthesis of hydroxycinnamic acid amides and flavonoids increased along with the change in the content of metabolites. These metabolites are known to be associated with antimicrobial activity and cell wall thickening (the latter helps prevent pathogen spreading in plant tissues from the site of infection and is considered a defensive reaction) [26-28].

In field conditions, metabolic changes in plants are due the combined action of various environmental factors. Thus, field trials are of an increased interest. In potato lines contrasting in resistance to phytophthora a comparative metabolic profiling of leaves by nuclear magnetic resonance (NMR) method with PCA data processing revealed clear clustering of samples collected during the onset of symptoms in susceptible forms [29]. Component loading showed that fatty acids, malate and rutin reaching higher levels in leaves of resistant varieties are resistance associated. Higher amount of succinate, by contrast, is characteristic of susceptible lines. Projection on the latent structures (PLS) method was also used to identify the relationship between the metabolite profile and the field resistance to the pathogen. The model developed for several varieties showed a high correspondence between the lesion described by the areas under disease progress curve (AUDPC) and the metabolite profile. VIP (variable importance in the projection) values showed that malate, rutin and sucrose play the greatest role in predicting the degree of the disease severity, the sucrose content positively correlates with AUDPC, and malate and rutin correlates negatively [29]. Metabolic analysis of samples collected in different stages of late blight led to the assumption that this approach can be reliable only when the profiles are used to compare dynamics over the course of infection. PCA method showed that the metabolite profiles of the samples collected at 12-day intervals are very different, and these differences are stronger than the inter-varietal variations. The authors indicated sucrose and malate as marker metabolites. Enzyme analysis of L-malate level to assess its relationship with late blight resistance confirmed the perspectives of metabolomics in assessment of plant resistance to pathogens and search for biomarkers of resistance [29].

A more detailed dynamic changes in the metabolism of potato tubers was studied in the unstable AC variety Novachip [30]. Analysis of polar and non-polar extracts revealed a total of 106 metabolites of which 95 were identified. Of these, 42 were attributed as pathogen-dependent, since their content significantly altered during late blight infection. The group of amino acids, including the precursors of secondary metabolites involved in defense against the pathogen, was subjected to the strongest changes. In Caesar and AC Novachip potato varieties having different resistance to phytophthora the metabolite profiling of tubers and leaves by GC-MS method revealed 77 compounds the content of which changed as the disease progressed [31]. Indole-3-acetonitrile, 3-hydroxybutyrate, D-mannitol, dihydrocoumarin and propionate were considered the protective metabolites, as their levels were much higher in more resistant variety Caesar. The spectrum of these compounds in the tubers and leaves is somewhat different, which may, according to the authors, indicate the features of the defense mechanisms in different organs. The authors also discuss prospects of new methods of metabolite analysis (e.g., NMR) in potato breeding for resistance [31].

Another paper reports on a close relationship of transcriptome and metabolome studies. RNA sequencing performed for the leaves of two resistant (F06025 and F06037) and one sensitive (Russet Burbank) potato genotypes revealed differences in the expression of 4216 genes in P. infestans infection [32], some of which encode enzymes involved in different metabolic pathways. Increased expression of these genes in resistant lines led to the accumulation of phenylpropanoids, flavonoids, alkaloids and terpenoids considered as protective

metabolites.

Rhizoctoniosis. Potato diseases caused by fungus *Rhizoctonia solani* attack the underground parts of the plant causing black scurf on stolons and tubers [33], which significantly decreases yielding [34]. A metabolite analysis by FT-ICR/MS (Fourier-transform ion cyclotron resonance) coupled with GC-MS method revealed peculiar metabolic pattern for infected potato shoots [1]. PCA analysis of 270 identified metabolites found differences between intact and *R. solani*-infected plants. PLS method also showed a clear difference between the metabolomes of healthy and affected seedlings. Mapping of infection associated alterations detected quantitative changes in the metabolites which are involved in 40 biosynthetic pathways and biochemically linked to 107 enzymes encoded by 222 genes [1]. Further search for marker metabolites indicated an increase in the activity of the mevalonate and deoxyxylulose phosphate pathways which led to activation in the biosynthesis of sesquiterpene pseudo-alkaloids (phytoberin, phytoalexins rishitin and solavetivon) and steroidal alkaloids with solasodine and solasodinin as aglycones. In infected seedlings, the content of most carboxylic acids, e.g. citramalate, oxalate, gluconate and α-keto-D-gluconate, was higher but pools of glucuronic and galacturonic acids, the cell wall components, decreased. In infection, the concentrations of substances potentially involved in systemic acquired resistance (SAR) mechanisms and hypersensitivity response (HR), such as azelaic acid and oxalic acid, were higher. The content of proteinogenic amino acids, except for pyroglutamic acid, was significantly lower in infected seedlings. Along with this, the pool of sugars in the infected shoots changed, viz. the content of D-fructose and myo-inositol decreased whereas the amount of other carbohydrates rose. The pathogen also influenced phenolic compounds in infected plant tissues, that is, the amounts of α-tocotrienol and ferulic acid increased while chlorogenic acid concentration decreased. At the same time, the content of amides of phenylpropanoids bound to the cell wall, in particular, N-feruloyl-tyramine, increased while the N-feruloylputrescine decreased [1].

Powdery scab. The disease of potato tubers and roots caused by *Spongospora subterranea* (Wallr.) Lagerh dramatically reduces plant productivity and tuber shelf life, which leads to significant economic damage. Root exudates in soil initiate germination of resting spores of the pathogen. Metabolomic analysis was used to identify marker compounds necessary for spore germination [35]. HILIC (hydrophilic interaction liquid) chromatography coupled with MS which mainly analyzes hydrophilic root exudates identified 24 low molecular weight compounds (mostly amino acids). Comparison of exudates of different potato varieties (Agria, Gladiator, Russet Burbank, and Iwa) revealed some peculiar features depending on the resistance of the variety to the pathogen [35

Thus, recent data indisputably show that different methods of metabolite analysis are good tools to study both progression of viral and fungal infections and resistance to viral and fungal diseases in plant species and varieties. Metabolite profiling of resistant potato varieties will facilitate potato breeding due to the use of biochemical markers.

Insect pests. The Colorado potato beetle (*Leptinotarsa decemlineata* Say), eating potato leaves, causes up to 30-50 % of crop losses [36]. Glycoalkaloids are a class of toxic metabolites of plant tissues, including potatoes, which protect plants from being eaten by herbivores. The main glycoalkaloids of *Solanum tuberosum* are solanine and chaconine [10], and the range of glycoalkaloids in other species of the *Solanum* genus is much wider [37, 38]. The leaf glycoalkaloids leptin and α-tomatin reduce the biomass eaten by adult beetles and increase the mortality of preimaginal larvae [39, 40]. In addition to alkaloids, many other metabolites are involved in

plant defense against Colorado potato beetles. For example, macrocypins, inhibiting cysteine proteases, interfere with the alimentation of herbivores [41]. Esters of some fatty acids and sugars are toxic [42], as well as sesquiterpenes [43].

Ultra high performance liquid chromatography coupled to quadrupole time flight spectrometry (UPLC-qTOF-MS) method used for leaf metabolite profiling in plants of potato wild species (*S. tarijense*, *S. oplocense*, *S. piurae*, *S. acroglossum*, *S. chomatophilum*, *S. paucissectum*) and varieties (*S. tuberosum*) with different Colorado potato beetle resistance elucidated the biochemical mechanisms underlying plant tolerance [44]. A search for metabolites associated with this trait showed that only *S. tuberosum* produces glycoalkaloids containing solanine and chaconine, the trisaccharide glycones. In the studied wild species, glycoalkaloids have tetrasaccharides in the side chains. In *S. oplocense*, *S. paucisectum*, *S. chomatophilum* and *S. piurae*, dehydrocommersonine, tomatine and neotomatine determine Colorado potato beetle resistance. A number of other metabolites that are not related to glycoalkaloids, for example, hydroxycoumarin and other phenylpropanoids, found only in wild species are also resistance-associated [44].

Sucking insects, including aphids, are another group of pests. Along with damage to the plant integuments and consumption of metabolites, these insects may often be the viral infection vectors. All this significantly reduces the potato yield [45, 46]. Unfortunately, to date, there is only one paper [45] on metabolite study with aphids on potatoes. The peculiarity of the work is that the metabolite profiles in leaves of different age, under attack of aphids and during viral infection were compared by NMR in intact and GMO lines. It was shown that the metabolite patterns change in all cases but to different extents. With genetic modification, the strongest metabolic differences manifest themselves in young leaves. The sucrose content significantly lowers whereas phenols and malate levels rise. The number of aphids eating on the plant does not differ between intact and genetically modified lines but depends on the content of glycoalkaloids which include α-solanine and α-hakonin. In young leaves, the accumulation of these metabolites is higher, which reduces the number of insects.

Thus, the metabolome analysis allows us to characterize the marker metabolites, the change in which underlies the response to lesions by leaf-eating and sucking insects.

In general, we can conclude that metabolite profiling, subject to more accessible equipment, can be a key approach in estimation of potato resistance to biotic factors.

Abiotic factors. Plant growth is largely determined by environmental conditions, i.e. physical and/or chemical factors. The light spectral composition, pool of macro- and microelements, temperature and water regimes can both accelerate and slow down plant development. Mechanisms underlying plant resistance to abiotic stressors at the transcriptome and proteomic levels is in focus of researchers since the 2000s, but metabolome studies are not numerous.

Drought and osmotic stress. Global warming leads to climate change, including a significant effect on the rainfall amount and duration. This may lead to droughts, salinization or flooding of agricultural lands. Plant resistance to moisture deficiency is the most studied. Accumulation of osmolytes (amino acids, sugars, polyols, etc.), antioxidants (glutathione, ascorbic acid, etc.) indicates a change in the intensity and direction of metabolic pathways [47], and therefore metabolite profiling is a tool to study the adaptation of plants to moisture deficiency [48].

It is known that potato plants are quite sensitive to the lack of moisture [49]. Metabolic rearrangements and their differences depending on drought resistance were compared in leaves, tubers and roots of the five potato genotypes

[50]. In the study, the authors used both non-targeted and targeted approaches. They examined about 7000 compounds in the first case and 60 in the second case, including the primary (glucose, malate, proline, etc.) and secondary (carotenoids, phenolic compounds, etc.) metabolites. It was shown that differences in metabolic alterations occur between organs and between genotypes. Stress-induced metabolic changes specific to each clone were analyzed by the targeted approach. The processes in the tubers were the focus. Organ-specific changes showed 45 compounds. In a lack of moisture, the accumulation of more complex phenolic compounds (naringenin, rutin and umbelliferone) is initiated in the leaves, but not in the roots and tubers. On the contrary, the content of their precursors and intermediates of the phenylpropanoid pathway (e.g. phenylalanine, chlorogenic and other phenylpropanoic acids) increased in all organs and in all genotypes, but with different intensity. Another group of metabolites accumulated in tubers is amino acids, e.g. glutamine, leucine, isoleucine, tryptophan, etc. However . the sugar level in tubers was almost independent of the stress factor, i.e. the changes mostly concerned only inositol and sorbose [50)].

Comparing leaf metabolites in Andean potato (*S. tuberosum* subsp. *andigena*) varieties Negra Ojosa and Sullu led to similar findings. These local varieties are more resistant to drought than varieties of cultivated potato *S. tuberosum* subsp. *tuberosum*. Metabolic profiling revealed a more pronounced accumulation of trehalose, proline and GABA in the less resistant variety Negra Ojosa at moisture deficit [51]. Concentrations of hexoses and complex sugars was practically unchanged and did not differ in both varieties, but in plants of more stable Sullu variety the level of organic acids of the Krebs cycle was higher during drought, which may indicate greater mitochondrial activity and stability.

The goal of a large-scale study of 31 potato varieties [52] was to search for drought resistance markers at transcriptome and metabolic levels by RNA sequencing and non-targeted metabolomic GC-MS detection. The minimum set of markers was 20 genes and metabolites. These allow prediction of drought resistance even at very early stages of cultivation. Interestingly, some markers were associated with resistance not only to drought, but also to pathogens, which suggests a commonality of resistance mechanisms to biotic and abiotic stressors.

Thus, metabolomic profiling can be a tool for phenotyping genotypes with different drought tolerance and study their physiological and molecular adaptations [50, 51].

With the use of an osmotically active sorbitol compound [53], two genotypes most contrasting in drought resistance were selected in vitro among two *Solanum* species and 18 varieties of *S. tuberosum*. Comparison of their proteomes under stressful conditions revealed different intensity of protein degradation combined with altered redox status [54]. Targeted metabolomic profiling of polar compounds detected changes for 26 of 42 metabolites examined in these two genotypes [53]. The authors evaluated the ratio of the amount of these compounds in experimental and control plants. A drought-induced decrease in the accumulation of metabolites occurred in most cases. Such dynamics was typical for ascorbate, aspartate, succinate, etc. The proline alteration is the most significant. In stress, proline accumulation was typical of both genotypes, but it was 11.39 times higher in the resistant ones. The accumulation of glycine, phenylalanine and sucrose was also noted. The obtained data are consistent with the dynamics of a number of earlier studied compounds [55].

Let us turn to another study in which metabolomic analysis was applied to assess tolerance to moisture deficiency in transgenic potatoes with a constitutively expressed *Arabidopsis* gene for transcription factor DREB that is induced during dehydration (DRE-protein protein/C-repeat binding factor; dehydration-

responsive element). DREB/CBF proteins bind to a specific site in the promoter and regulate gene expression during drought and low temperature [56]. A non-target metabolomic approach detected 165 metabolites (113 were identified) and revealed differences in tubers between control and transgenic plants. The most essential rearrangements in transgenic plants (i.e. higher accumulation) were characteristic of compounds involved in glutathione and GABA metabolism, which, according to the authors, indicates the activation of adaptation mechanisms due to the expression of the DREBA encoding gene. The presence of this protein can have both direct effect by changes in gene expression and an indirect effect, for example, by initiation of the ethylene phytohormone synthesis [56]. It should be emphasized that transgenic lines did not accumulate toxic solanine and chaconine. That is, an increase in the stability of transgenic lines did not lead to deterioration in consumer qualities of tubers. Thence, metabolic analysis is sensitive enough to detect differences in constructed potato lines.

High temperatures. Temperature is one of the most unpredictable abiotic factors affecting the growth and yield of potatoes [57]. Given that the potato was originally domesticated in the highland regions of the central Andes of South America, it is not surprising that these varieties are most productive in the temperature range of 15-19 °C [58]. In the future, due to climate warming, global potato production may decrease by 20-30 % [59], therefore, selection for resistance to elevated temperatures is becoming more relevant.

Genomic and transcriptome studies lead to the conclusion that resistance to elevating temperature is regulated by a large number of genes, changes in the expression of which affect biochemical and physiological reactions [57]. This generates more interest in studying the adaptation mechanisms of potatoes to rising temperatures at transcriptome and metabolome levels simultaneously (60). Significant rearranngements are shown for 89 of 123 detected metabolites. When hyperthermia decreased, the content of most of them diminished, including amino acids and nitrogen-containing compounds, such as ethanolamine and putrescine. The accumulation of the latter decreased both in leaves and tubers. The same trends were observed for organic acids associated with the tricarboxylic acid cycle, and for carbohydrates. The decrease in fumarate and succinate concentrations was especially pronounced in tubers. In leaves, a decrease in fructose, galactose and their phosphorylated forms was more intensive. A tuber specific change of metabolome with an increase in temperature affected the content of alcohols (decrease in the amount of sorbitol and mannitol, increase in inositol), lipid composition (higher saturation of fatty acids, greater amounts of C28-C30 fatty acids) and higher level of fatty alcohols, especially phytosterols.

The revealed metabolite rearrangements completely correlate with changes in the expression of genes encoding the enzymes of the corresponding biochemical cycles, which indicates the possibility of using the metabolomic approach to analyze biochemical rearrangements in potato plants influenced by abiotic stressors [60].

The available data indicate that the metabolomic approaches are sensitive enough to assess the effects of exposure to stressors directly related to cell dehydration (these changes mainly concern the composition of sugars and amino acids). The further studies will find out how metabolic rearrangements may be simultaneously influenced by several factors, for example, lack of moisture in combination with higher temperature.

Plant resistance to biotic agents is determined by immunity, including recognition receptors, the pathogen-associated molecular patterns, PAMPs, or microbe-associated molecular pattern, MAMPs) and effector-triggered immunity [13]. Resistance to abiotic factors is also based on highly specific recognition of

the stressor and on adaptive response, which can be divided into nonspecific and specific. A huge number of studies have shown that the basis of resistance is the activation of genes, including those responsible for quantitative traits (QTLs) [61]. In the present review, we have shown, using the example of potatoes, that the various metabolic reorganizations are dynamic processes depending on the type and strength of the stressor, as well as on the plant genotype. Adaptation to biotic and abiotic factors is provided by the accumulation of secondary metabolites and changes in the balance of amino acids and sugars.

Creating stress resistant potato varieties includes interspecific hybridization with wild-growing forms, cell engineering and genetic transformation [62]. Currently, along with genomic and proteomic technologies, metabolomic methods are being actively used in studying plant resistance and phenotyping wild and cultivated species, hybrids, varieties, and transgenic forms [8]. Recent strategy proposed to improve potato growing is based on the production of seeds of heterotic diploid hybrids (true potato seeds, TPS) [63]. Note that each of these approaches has its advantages and limitations.

In fact, all the data on potato metabolome have been obtained in the study of the vegetative organs of plants. The metabolome features of generative organs are currently not studied, there is completely no information on the metabolome profiles of CMS forms which are necessary for heterosis selection [64]. The available data indirectly indicate significant rearrangement in the metabolite profiles during the formation of pollen in *Solanacea* family, especially under the influence of elevated temperatures [65], which confirms the relevance of this trend in the study of potato metabolome.

So, metabolomic profiling becomes an integral part of basic research elucidating mechanisms of resistance to adverse biotic and abiotic factors. In addition, metabolomic analysis, after standardization of the analytical methods and data processing, can become a key element, and over time, the main technique to monitor potato plants in collections, created varieties and hybrids. There is no doubt that such studies are promising for the phenotyping potato genotypes, as well as for identifying forms that are resistant to various adverse effects.

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