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GENERAL BIOLOGY

Molecular Heterogeneity of Lectins in Wheat Seedlings under the Action of Stevioside and Heavy Metals

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Abstract—The effect of the diterpene glycoside stevioside and high concentrations of heavy metals on the molecular heterogeneity of lectins was studied in seedlings of Kazanskaya 560 winter wheat cultivar. Stevioside induced the emergence of a new 45-kDa lectin. Cultivation of wheat seedlings in $CdSO_4$ and $ZnSO_4$ solutions resulted in the emergence of the protein with Mr = 88 kDa. We detected the presence of both lectins in seedlings during combined treatment with stevioside and heavy metals.

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At present, substances isolated from plants are an extremely promising basis for the design of new biologically active compounds exhibiting a pronounced anti-stress effect. Their use makes it possible to increase the plant stability and productivity under the action of stress factors [1].

Among these compounds, kaurene derivatives, which include tetracyclic diterpenoid steviol (an aglycone of glycosides obtained from *Stevia rebaudiana* Bertoni grass extract), attract a special attention. Stevioside is one of glycosides of this plant.

Previously [2–4], we established that stevioside at a concentration 10^{-8} M activated the growth and increased the frost resistance of winter wheat plants, decreased a negative effect of cadmium and zinc on the plant growth, as well as changed the activity of lectins. Since the role in regulation of plant growth and development and performing a protective function under biotic and abiotic stressful conditions have been established for lectins [5–8], the aim of the present study was to clarify the molecular heterogeneity of lectins in the cell wall of winter wheat plants during treatment with stevioside and heavy metals (HM).

The roots of the Kazanskaya 560 cultivar of *Triticum aestivum* L. winter wheat seedlings were the object of the study. Stevioside was obtained from plant raw material (stevia) in the Arbuzov Institute of Organic and Physical Chemistry (Kazan Scientific Center, Russian Academy of Sciences, Kazan, Tatarstan, Russia). The plants were grown under laboratory conditions in cuvettes containing tap water, under illumination of 10 000 lx, photoperiod of 12 h, and temperature of 23°C for 7 days. In experimental variants, the plants grew in a stevioside solution (10^{-8} M). The optimal stevioside concentration was selected in preliminary experiments [2]. Isolation of cell wall lectins was conducted according to the methods described previously [9]. Gel filtration chromatography was used to analyze the molecular heterogeneity of lectins. Sephadex G-150 fine (Sigma-Aldrich, United States) was used as a sorbent. In the collected fractions (1 mL each), the protein content (by the Lowry method) and the activity of lectins (by means of the hemagglutination reaction with erythrocytes of blood group O) were determined [10].

As can be seen from Fig. 1, the addition of stevioside to the incubation medium resulted in a decrease in the activity of the cell wall lectins. To clarify this question in more detail, we studied the possible differences in molecular weight of the cell wall lectins.

In the control variant (cultivation without stevioside), we found lectin activity in the proteins with molecular weights of 93, 77, 63, 36, and 19 kDa among the protein fractions obtained after chromatography (Fig. 2).

The 36-kDa protein can be presumably the classical wheat lectin, wheat germ agglutinin (WGA). It is known that WGA can be in the space between the plasmalemma and cell wall (in addition to cytoplasm). This agglutinin (rapid accumulation of which occurs under different unfavorable conditions) is an excreted protein [11].

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