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Haemolymph nutrient homeostasis in the Tenebrio molitor larvae affected by Solanaceae glycoalkaloids

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Introduction

Materials and methods

Glycoalkaloids (GAs) are produced by many plant species and have high biological activity. In plants, they act as natural protectants against pathogens and herbivores. The studies of GAs in insects are focused mainly on the lethal and sublethal effects. There are only a few studies concerning the mechanisms of their action in these organisms and, thus, the activity of these compounds remains largely unknown. The aim of our studies was to determine whether GAs have impact on insect **metabolism.** The metabolism is tightly connected with nutrition, and as that GAs may be ingested together with nutrients, it seems that they also should affect metabolism (Spochacz et al. 2018).



To check if tested compounds change metabolic activity of insect cells thus, have an impact on availability of nutrients for other tissues, we analyzed the levels of glicerol, free amino acids and carbohydrates in insect haemolymph. In the research, three GAs in concentrations 10^{-8} and 10^{-5} M were tested: solanine, chaconine and tomatine. The content of nutrients was determined in the haemolymph of *T. molitor* larvae (Fig. 1a) 24 hours after injection of 2 μ l of the tested GAs. In control, insects were injected with physiological saline. The assays were performed by RP-HPLC technique (Fig. 1b).



Fig. 1 a) The larvae of *T. molitor* beetle, b) The HPLC equipment.



Results and conclusions

The analysis revealed the effect of the tested compounds on glycerol, amino acids and carbohydrates (Fig. 2) content haemolymph. Generally, GAs increase the level of most metabolites in haemolymph. Statistically significant differences in the concentration of amino acid valine and carbohydrate glucose are observed after the injection of the higher concentrations of tested compounds. Almost all GAs significantly raise the level of sorbitol in haemolymph. On the other hand, decreased level of sucrose as a result of solanine and chaconine injection (10⁻⁵ M) compared to the control is noted. These changes may be the result of GAs detoxification which requires high energy inputs.

Obtained results suggest that GAs have impact on insect physiology and influence on nutrient balance in haemolymph. Detailed determination of their mode of action in insects is extremely important, because it could enable projecting new biopesticides, as GAs show broad insecticidal activity (Chowański et al. 2016). Taken together, this research provides important insights into GAs action in insect tissues.

Sorbitol



Fig. 2 The concentration of glycerol, amino acids and carbohydrates in *T. molitor* haemolymph after injection of tested GAs (mean values ± SEM). *** Statistical significance at $p \le 0,001$, ** $p \le 0,01$, * $p \le 0,05$, Student's *t*-test (for glycerol, prolina and mannitol), Mann Whitney test (for the rest).

<u>References:</u> Spochacz M. et al. 2018, *Sublethal Effects of Solanum* nigrum Fruit Extract and Its Pure Glycoalkaloids on the Physiology of Tenebrio molitor (Mealworm), Toxins, 10(12), 504; Chowański S. et al. 2016, A Review of Bioinsecticidal Activity of Solanaceae *Alkaloids*, Toxins, 8(3):60.





