Abstract

Insects have developed many strategies of defence against predators in the course of evolution. The evolutionarily oldest and most widely used type of defence is chemical defence, followed by acoustic or optical defence. However, many species of insects use simultaneously multiple types of warning signals, which affect different sensory receptors of the given predator. Such a complex method of warning signals is called multimodal method. It may consist of a combination of simultaneous chemical and optical signals, or a combination of acoustic and optical signalling. The combination of chemical and optical signalling used against a predator is probably the most common form of multimodal signalling. The presented work deals with the analysis of biologically active substances, which participate in the defence mechanisms of a widespread species of insects – true bugs (Heteroptera).

Pterin derivatives represent a large group of natural compounds derived from pteridin, bicyclic heterocycle, and they are found in virtually all living organisms from bacteria to vertebrates. In insects, they primarily serve as pigments, resulting for example in striking coloration of cuticles of Heteroptera. The first part of the dissertation was focused on identification and quantification of pterin derivatives in cuticles of selected species of true bugs. The method of capillary zone electrophoresis with UV detection and hydrophilic interaction chromatography in combination with tandem mass spectrometry was developed for the purpose of separation and quantification of ten pterin derivatives (L-sepiapterin, 7,8-dihydroxanthopterin, 6-biopterin, pterin, D-neopterin, isoxanthopterin, leukopterin, xanthopterine, erythropterin, pterin-6-carboxylic acid). Both methods were validated in terms of conventional validation parameters - linearity, accuracy, precision, limit of detection and quantification etc. The methods were used to identify and detect distribution of pterin derivatives in selected species of true bugs.

True bugs have very well developed scent glands, which —when irritated - produce large amounts of highly odorous substances functioning as chemical protection against predators. The second part of this work dealt with appropriate strategies of collection of volatile secretions of true bugs. New, non-invasive methods of collection of volatile secretions of true bugs have been designed, with their subsequent separation by means of gas chromatography with mass spectrometry. Pre-concentration of secretion samples was performed by adsorption on SPME fibre. Three types of SPME fibre were tested for the reason of high complexity of the secretion samples: non-polar polydimethylsiloxane (PDMS), polar polyacrylate (PA) and bipolar

divinylbenzene/carboxen/polydimethylsiloxane (DVB/CAR/PDMS). The entire optimization procedure was performed using a factorial design - RSM (response surface methodology).

Modern capillary fluorinated phase Rtx-200 was used for GC separation providing good selectivity for substances from a wide range of polarities. The elaborated methodology was applied to analyse defensive secretions of males and females in selected species of true bugs.