Abstract

Among messenger molecules neuropeptides represent the largest group with respect to their functional and structural diversity. Neuropeptides can act as transmitters, modulators and hormones and play an important role in the regulation of virtually all physiological processes like homeostasis, development, nutrition, and reproduction. Furthermore some neuropeptides are also known to adopt other functions such as growth factors, toxins or antimicrobial agents.

Insects are favoured model organisms in the investigation of neuronal, endocrine and neuroendocrine regulation mechanisms due to their simple rearing conditions, manipulability and medical, economic and ecologic importance. The neuropeptidome of insects was extensively studied in the past decades with mass spectrometry representing the leading method. However, larger peptides or even proteins, which for the most part contain cysteine bridges or other modifications such as phosphorylations, were missing in most studies.

The work in hand focuses on such peptides and utilizes novel methods in the field of MS-based neuropeptidomics to overcome limitations of analytical setups. It also challenges the small sample amounts given the size of insect organs. The developed workflows not only include sample preparation and enrichment, peptide modification and enzymatic cleavage, but also employ bioinformatics. The tools applied has enhanced the scope of studies based on matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) and has enabled the identification and discovery of formerly inaccessible neuropeptides and protein hormones by means of MALDI-TOF MS.

The present work demonstrates the identification of phosphorylated pyrokinins encoded by the *capa* gene, *de novo* sequencing of polypeptide dimers encoded by *akh* genes, the discovery of a previously unknown cystine-knot containing agatoxin-like peptide (ALP) and the identification and *de novo* sequencing of exceptionally large peptides. The study covers the full precursor of the ovary maturation parsin (OMP) / corticotropin-like releasing factor diuretic hormone (CRF-DH), the protein hormone neuroparsin as well as other peptides and proteins in the neuroendocrine system.

The discovery of these peptides and proteins in neuroendocrine tissues of well studied model organisms like *Periplaneta americana* and *Apis mellifera* encourages scientists to revisit the neuropeptidome of other popular insect model organisms and to evaluate the functions of these compounds.