

P043

Comparative venomic characterization of four medicinally important Turkish vipers (*Macrovipera lebetina*, *Montivipera xanthina*, *Vipera ammodytes*, *V. kaznakovi*)
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Introduction: Exploring venom proteomes is of great interest due to their potential as sources of bioactive peptides and proteins with novel pharmacological properties.

Objectives: Our aim is to characterize the venom proteome of four Turkish vipers in order to evaluate their potential as a source for bioactive molecules, as well as to determine specific venom variations.

Materials and Methods: Snakes were collected in various regions of Turkey and pooled venom samples were freeze-dried after extraction. Bottom-up proteomics experiments were carried out on a Thermo Orbitrap XL mass spectrometer (MS) equipped with an Agilent 1100 HPLC system. Crude venom proteins were separated by reverse phase chromatography (RPC) using C18 column, under increasing acetonitrile gradient. Each fraction were collected and evaporated in a vacuum centrifuge. Selected fractions were loaded onto 12% SDS-PAGE gels and stained with Coomassie Brilliant Blue. Protein bands were excised and in gel digestion was performed with trypsin. The resulting peptides were submitted to the same LC-MS/MS system using C18-RPC for prior peptide separation. MS/MS fragmentation spectra were obtained for selected peptides using CID and HCD methods in combination. *De novo* peptide sequences were obtained either by manual data interpretation or by Peaks *de novo* software tool. Thereafter, sequences were submitted to BLASTP to identify the proteins.

Results: We identified phospholipase A₂ (PLA₂), metalloproteinase (SVMP), serine proteinase (SVSP), cysteine-rich secretory protein (CRISP), disintegrin (DISI), c-type lectin (CLP), L-amino acid oxidase (LAAO), vascular endothelial growth factor (VEGF), nerve growth factor (NGF), proteinase inhibitor and bradykinin-potentiating peptides (BPP) totally in all venoms. However, distribution and abundance of these proteins, which were calculated, based on the corresponding peak areas of UV₂₁₄-RPC among studied venoms shows variation and is demonstrated in the Figure 1.

Conclusions: In this work, the first detailed proteomic insight into the Turkish viper venoms was achieved, including totally unexplored venoms like *M. xanthina* and *V. kaznakovi*. Our results show that these venoms contain biotechnologically important proteins and peptides that will be a guide for our further studies on the purification of bioactive proteins. Furthermore, the determination of the compositional variations of different Turkish viper venoms is of great importance for developing much more potent antivenoms and our comprehensive study gives the first insight into the venom variation of Turkish vipers.

References: Calvete JJ, Sanz L, Angulo Y, Lomonte B, Gutierrez JM (2009) Venoms, venomics, antivenomics. FEBS Letters, 583: 1736-1743.

Figure 1: Comparative protein/peptide compositions of *Macrovipera lebetina*, *Montivipera xanthina*, *Vipera ammodytes* and *V. kaznakovi*

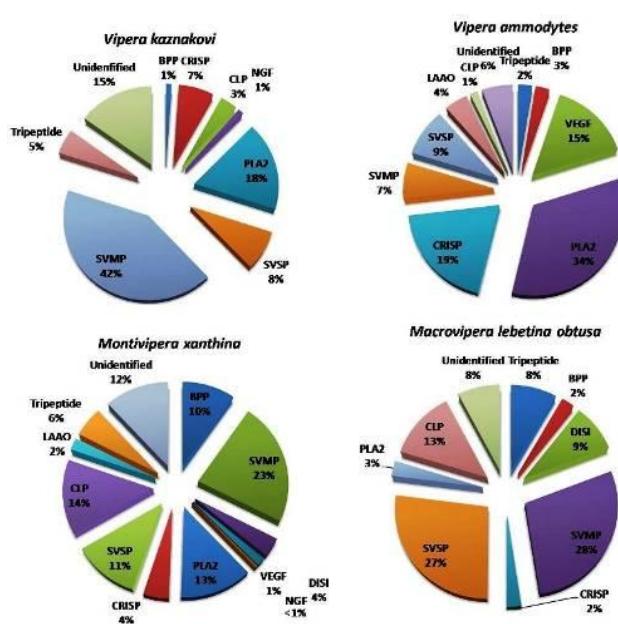
Figure 1

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