High-throughput epitope profiling of snake venom toxins
unveiling the complexity of antigen-antibody interactions of antivenoms

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Publication date:
2016

Document Version
Publisher's PDF, also known as Version of record

Link back to DTU Orbit

Citation (APA):
High-throughput epitope profiling of snake venom toxins – unveiling the complexity of antigen-antibody interactions of antivenoms

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Introduction
Insight into the molecular details of polyclonal antivenom antibody specificity is a prerequisite for accurate prediction of cross-reactivity and can provide a basis for design of novel antivenoms1. In this work, a high-throughput approach was applied to characterize linear elements in epitopes in 82 toxins from four African mamba and three neurotoxic cobra snakes obtained from public databases.

Studying linear epitopes using peptide microarrays

Key residues for antivenom toxin recognition

Antivenoms antibodies bind to functional sites of toxins

Conclusions
Custom-designed high density peptide microarray technology enables parallel automated identification of linear elements of epitopes in snake neurotoxins.

Trend: antivenom antibodies recognize and bind to epitopes at the functional sites of toxins.

Perspectives
Determination of linear elements in snake venom toxin epitopes may provide the basis for:
• Explaining the molecular basis of antivenoms para-specificity
• Guiding next-generation antivenoms based on DNA immunization and immunization with synthetic epitope strings

Figure 3. Structural presentation of B-cell epitope analysis: (A-D) Short neurotoxin 1 (P01410) from D. polysoga as an example of a type 1 α-neurotoxin. Structure built upon: (B-C) Fasciculin-2 (P01278) from D. argenticeps as an example of a fasciculin. The Fasciculin-2 is co-crystallized with the human acetylcholinesterase enzyme. Structure built upon: (D-F) Tryx P3-5 (P01396) from D. polylepis as an example of a δ-neurotoxin. The Tryx P3-5 (P01396) is a Fab from D. polylepis Opgi (P01396), which is designed to mimic the Cβ subunit of α-neurotoxins. (A) Antivenoms antibodies bind to functional sites of toxins. (B) Type 1 α-neurotoxins.

Figure 2. Examples of B-cell epitope analysis: Types 1 and 2 α-neurotoxins and dendrotoxins recognized by the SAIMR polyvalent antivenom. The heat profile above each sequence represents the average score of peptides containing a given peptide. The color background represents the average amino acid substitution effect. Where: 12-mer peptide covering a given residue passed the epitope-threshold, the residue is colored grey. Dark purple indicates that a residue is of particular importance for antibody recognition.

Figure 1. (A-D) Venn diagrams of peptides classified to bind antivenom antibodies for each pair of experiments conducted with the same antivenom in two different dilutions: (A) SAIMR Polyvalent Snake Antivenom, (B) VLS Opolysoga, and (C) VLS Central Africa. (D) Venn diagram of peptides classified as binders for each antivenom. Only peptides identified in both experiments with each antivenom, corresponding to the overlaps in Venn diagram in part A-D, are included.

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Acknowledgement
We would like to thank Morten Nielsen for scientific discussion and the Novo Nordisk Foundation for financial support (grant number NFF1300030013)