

Proteomic analysis and lethality of the venom of *Aegaeobuthus nigrocinctus*, a scorpion of medical significance in the Middle East

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Abstract

The scorpion *Aegaeobuthus nigrocinctus* inhabits areas in Turkey and the Levant region of the Middle East where severe/lethal envenomings have been reported. Previous research indicated its extreme venom lethality to vertebrates and distinct envenomation syndrome. We report on the composition of *A. nigrocinctus* venom from Lebanese specimens using nESI-MS/MS, MALDI-TOF MS, SDS-PAGE and RP-HPLC. Venom lethality in mice was also assessed ($LD_{50} = 1.05$ (0.19–1.91) mg/kg, i.p), confirming *A. nigrocinctus* venom toxicity from Levantine populations. Forty-seven peaks were resolved using RP-HPLC, 25 of which eluted between 20 and 40 % acetonitrile. In reducing SDS-PAGE, most predominant components were <10 kDa, with minor components at higher molecular masses of 19.6, 26.1, 46.3 and 57.7 kDa. MALDI-TOF venom fingerprinting detected 20 components within the 1,000–12,000 m/z range. Whole venom ‘shotgun’ bottom-up nLC-MS/MS approach, combined with in-gel tryptic digestion of SDS-PAGE bands, identified at least 67 different components belonging to 15 venom families, with ion channel-active components (K^+ toxins (23); Na^+ toxins (20); Cl^- toxins (2)) being predominant. The sequence of a peptide (named α -KTx9.13) ortholog to *Leiurus hebraeus* putative α -KTx9.3 toxin was fully determined, which exhibited 81–96 % identity to other members of the α -KTx9 subfamily targeting Kv1.x and Ca^{2+} -activated K^+ channels. Chlorotoxin-like peptides were also identified. Our study underscores the medical significance of *A. nigrocinctus* in the region and reveals the potential value of its venom components as lead templates for biomedical applications. Future work should address whether available antivenoms in the Middle East are effective against *A. nigrocinctus* envenoming in the Levant area.