

Supplementary Materials: Novel Three-Finger Neurotoxins from *Naja melanoleuca* Cobra Venom Interact with GABA_A and Nicotinic Acetylcholine Receptors

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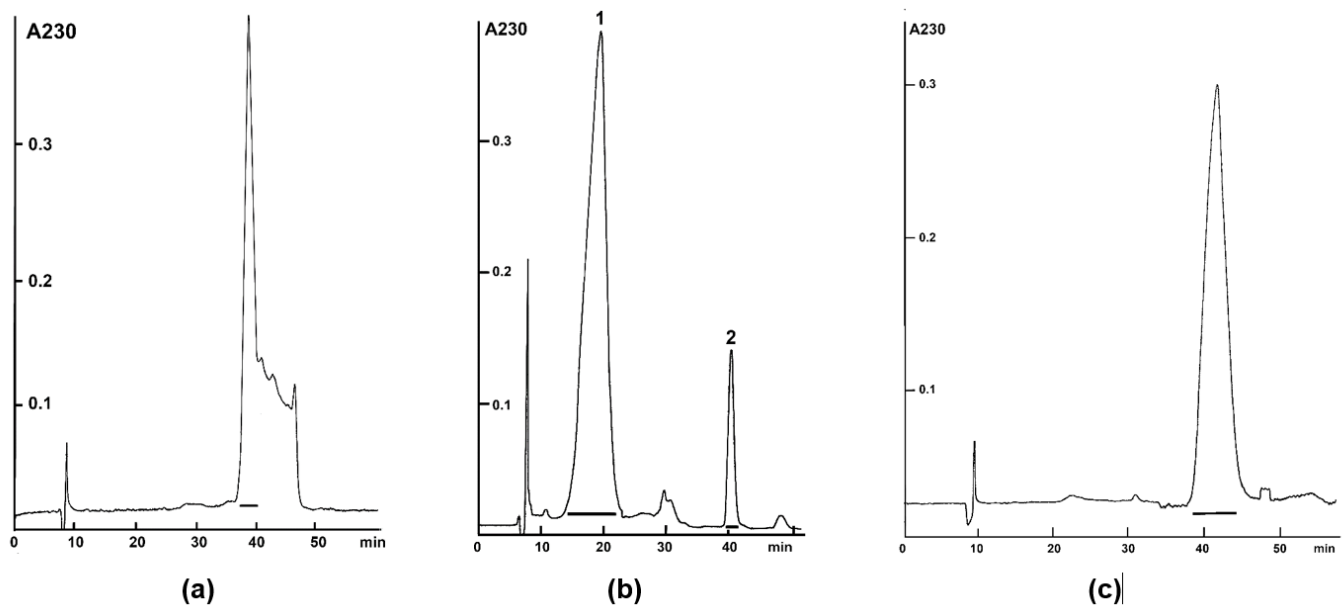
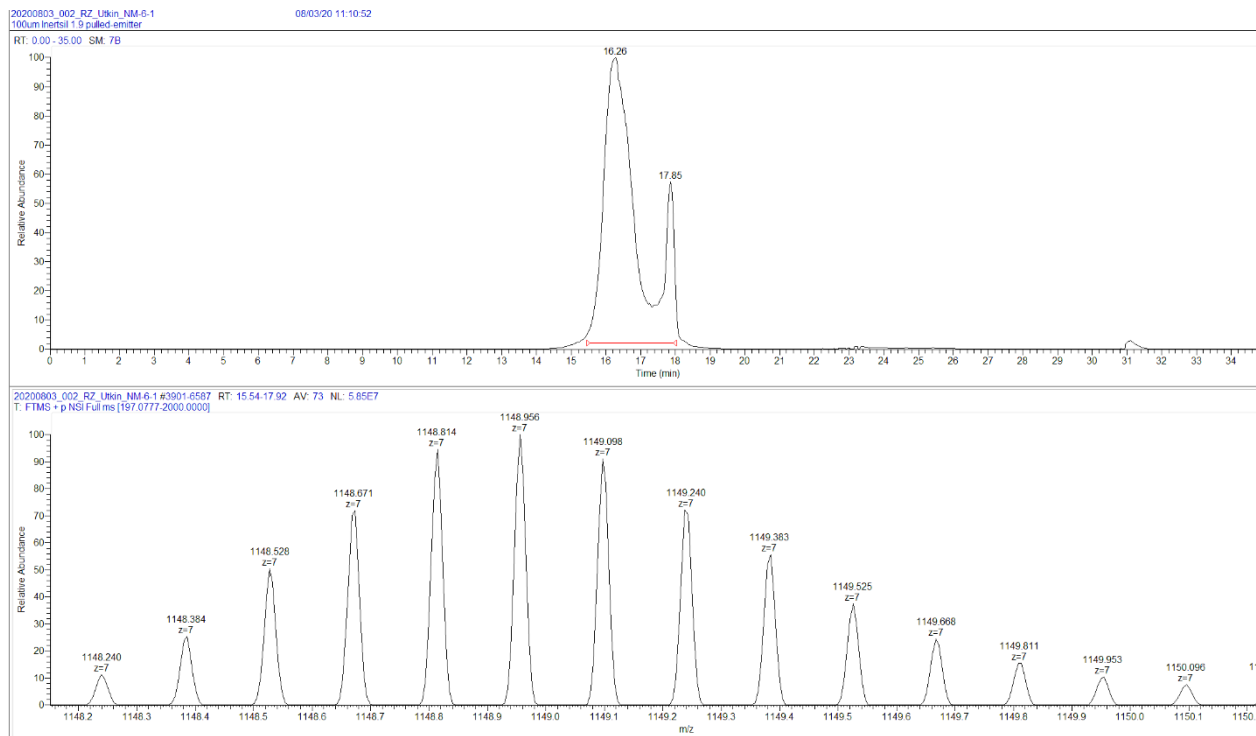
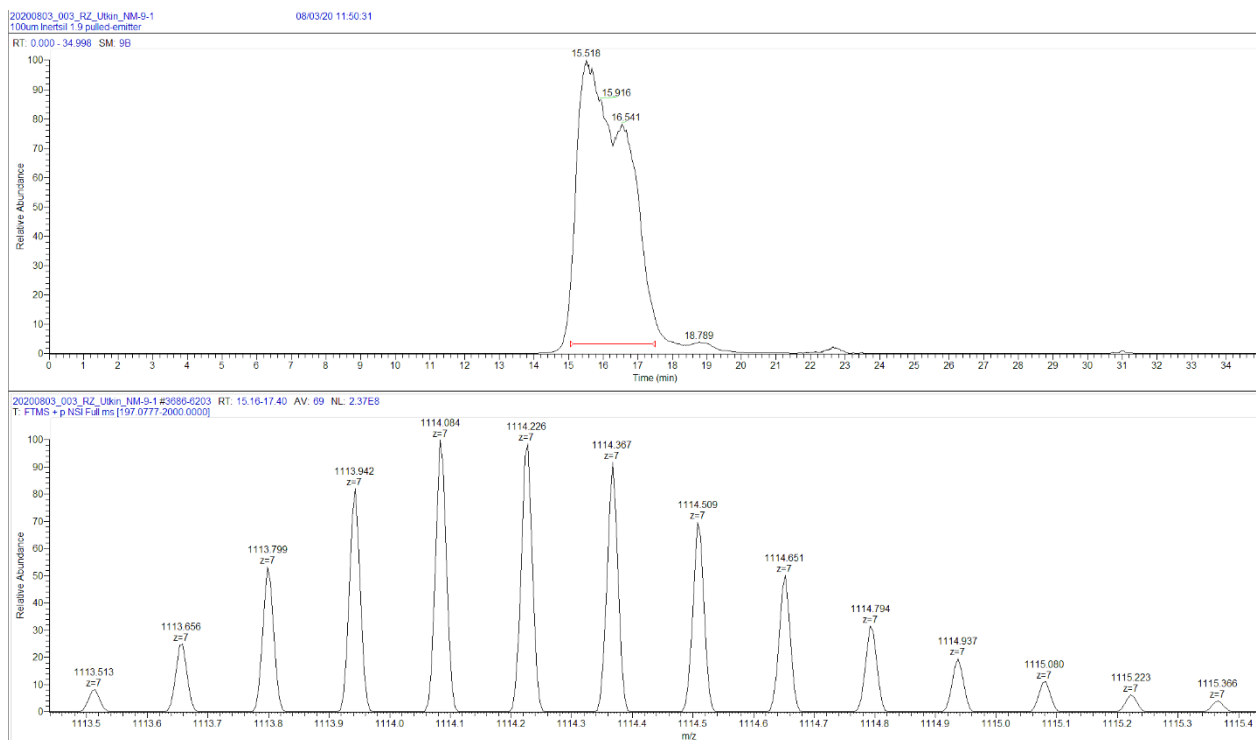


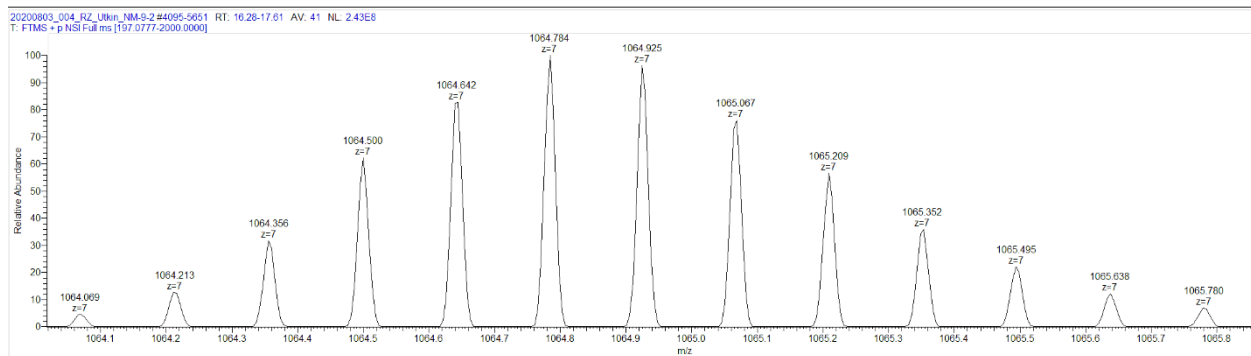
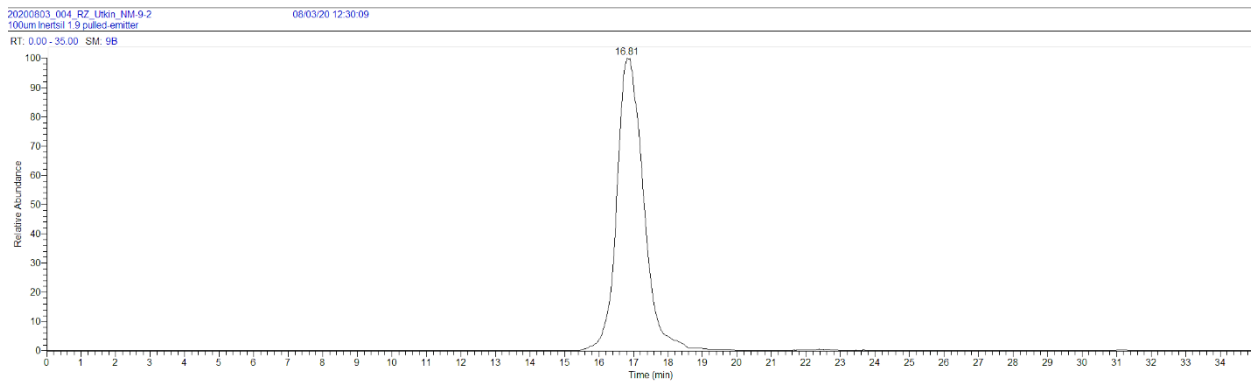
Figure S1. Separation of fractions 2 (a), 3 (b) and 4 (c) after ion exchange chromatography (Figure 1b) by reversed phase HPLC on Jupiter C18 column (10 × 250 mm, Phenomenex, Torrance, CA, USA) in a gradient of acetonitrile 20–35% in 60 min in the presence of 0.1% trifluoroacetic acid, at a flow rate of 2.0 mL/min. The horizontal bars indicate the fraction collected for Tx-NM2 (a), Tx-NM3-1 and Tx-NM3-2 (b), as well as Tx-NM4 (c).



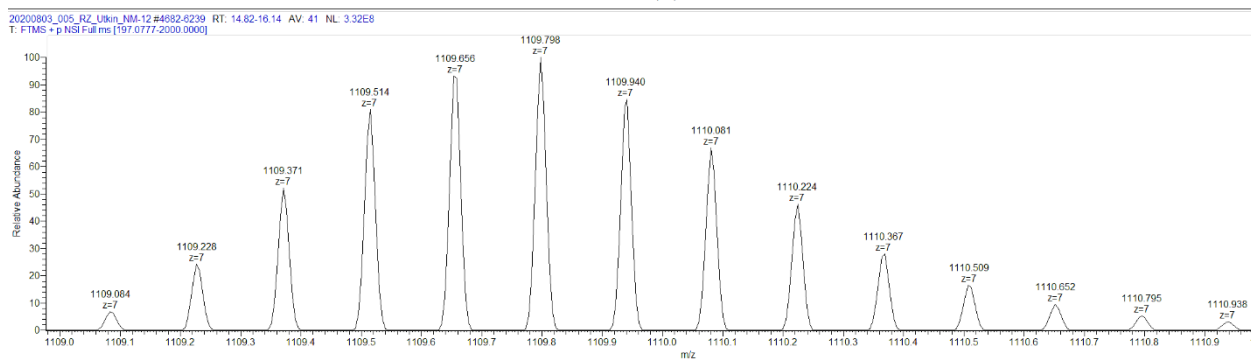
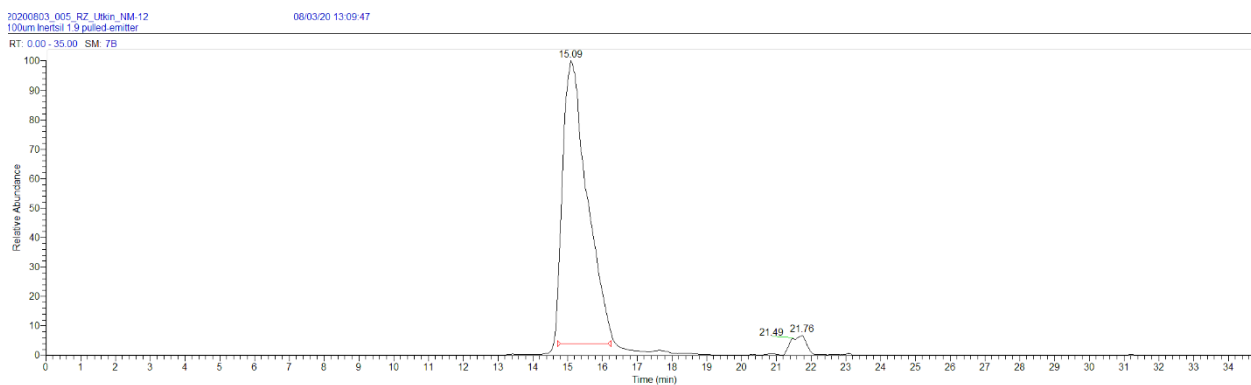
(a)



(b)



(c)



(d)

Figure S2. Extracted-ion chromatogram (XIC) from an LC-MS analysis and high-resolution spectra at $z = 7$ for toxins Tx-NM2, XIC at $m/z = 1148.5–1149.5$ (a); Tx-MN3-1, XIC at $m/z = 1113.5–1114.5$ (b); Tx-Nm3-2, XIC at $m/z = 1064.5–1065.5$ (c); and Tx-NM4, XIC at $m/z = 1109.5–1110.5$ (d). Each of two peaks observed for Tx-NM2 (a) and Tx-MN3-1 (b) give the same masses and represent toxin conformers. The signals with the lowest m/z ratio were used for the calculation of the monoisotopic mass by the following formula—protein mass = $(m/z \times z) - M_H \times z$, where z is charge state equal to 7; M_H , atomic mass of proton equal to 0.998 a.u.