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Title:	N-Terminal Region of <i>Vibrio parahemolyticus</i> Thermostable Direct Hemolysin Regulates the Membrane-Damaging Action of the Toxin
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Keywords:	<i>Vibrio parahemolyticus</i> Toxin Thermostable direct hemolysin (TDH)
Issue Date:	2019
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Abstract:	Thermostable direct hemolysin (TDH) of <i>Vibrio parahemolyticus</i> is a membrane-damaging pore-forming toxin with potent cytolytic/cytotoxic activity. TDH exists as a tetramer consisting of protomers with a core β -sandwich domain, flanked by an 11-amino acid long N-terminal region (NTR). This NTR could not be modeled in the previously determined crystal structure of TDH. Moreover, the functional implication of NTR for the membrane-damaging action of TDH remains unknown. In the present study, we have explored the implications of NTR for the structure–function mechanism of TDH. Our data show that the presence of NTR modulates the physicochemical property of TDH in terms of augmenting the amyloidogenic propensity of the protein. Deletion of NTR compromises the binding of TDH toward target cell membranes and drastically affects the membrane-damaging cytolytic/cytotoxic activity of the toxin. Mutations of aromatic/hydrophobic residues within NTR also confer compromised cell-killing activity. Moreover, covalent trapping of NTR, via an engineered disulfide bond, against the core β -sandwich domain also abrogates the cytolytic/cytotoxic activity of TDH. This observation suggests that an unrestrained configuration of NTR is crucial for the membrane-damaging action of TDH. On the basis of our study, we propose a model explaining the role of NTR in the membrane-damaging function of TDH.
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