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NaD1 forms oligomeric complexes with phosphatidylinositol to lyse cell membranes

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Cationic antimicrobial peptides (CAPs) are innate defense molecules produced by essentially all living species and represent a rich source of novel therapeutic molecules for the treatment of human diseases including cancer. Although several CAPs (mainly from amphibians and vertebrates) have been shown to exhibit direct toxicity against tumor cells, the underlying molecular mechanisms are not well understood. Nicotiana alata (ornamental tobacco) defensin 1, NaD1, is a Class II solanaceous plant defensin in the CAP family that exhibits antifungal activities. We have identified NaD1 as a potent molecule in killing mammalian tumour cells. Microscopy analyses have revealed that NaD1 induces cytolysis of tumor cells via the formation of large plasma membrane blebs. NaD1 was demonstrated to bind directly to phosphatidylinositol 4,5-bisphosphate (PtdIns(4,5)P2 or PIP2) resulting in disruption of cytoskeleton-membrane interactions and subsequent plasma membrane blebbing. To define the molecular basis of the interaction between NaD1 and PIP2, we determined the crystal structure of a NaD1:PIP2 complex. Strikingly, NaD1 forms a unique arch-shaped oligomer comprised of seven NaD1 dimers and 14 PIP2. The structure of the protein:lipid complex indicates that the presence of PIP2 is critical for oligomerisation of NaD1. Formation of NaD1:PIP2 oligomers was further confirmed by protein-protein crosslinking and transmission electron microscopy. Based on these data we propose that NaD1 is an innate pattern recognition molecule for PIP2 and forms a unique protein-lipid oligomeric complex that mediates permeabilisation of both microbes and tumour cells.

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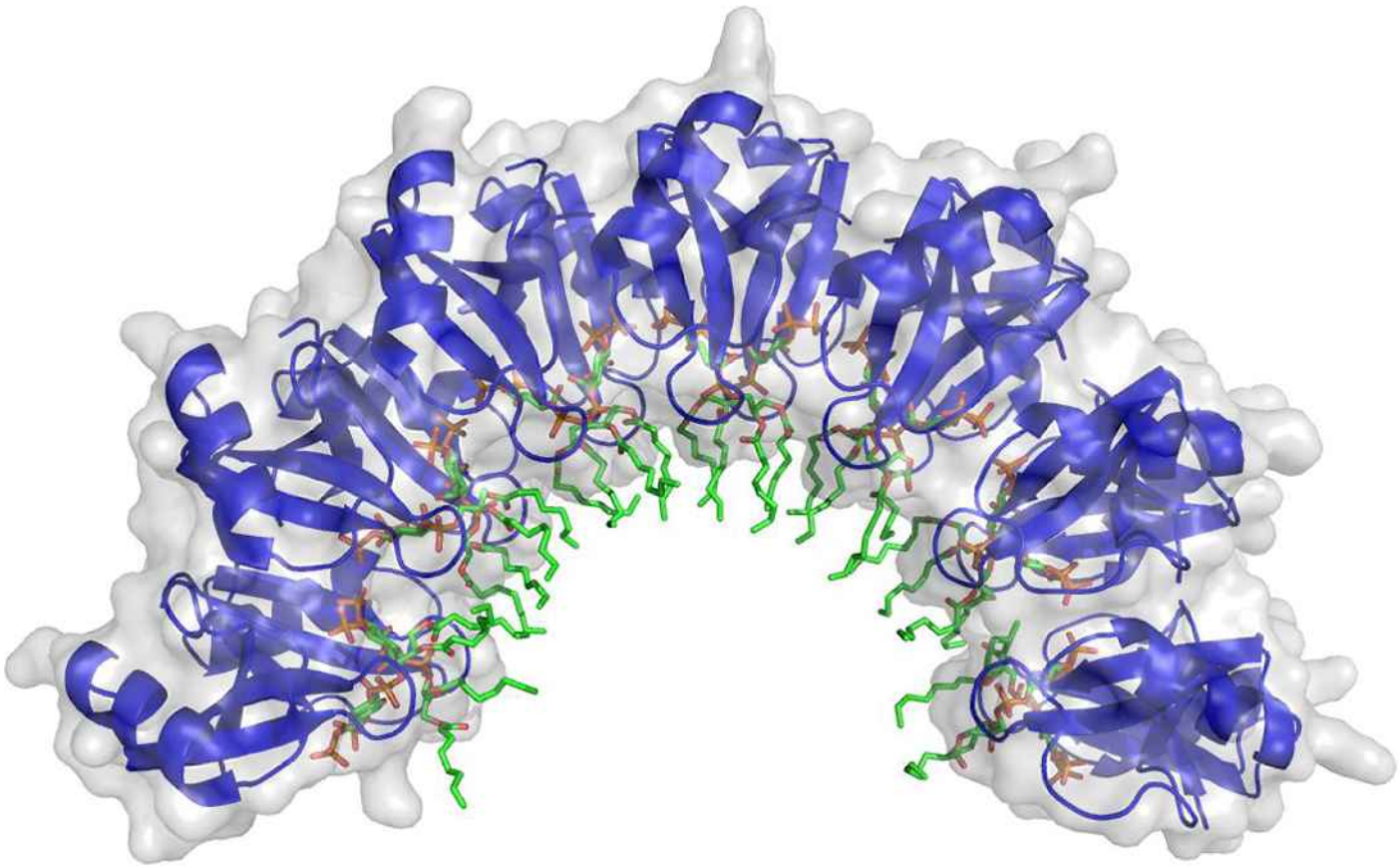

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