Lipid shape is a key factor in the synergistic reorientation of membrane-bound PGLa and magainin 2

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The antimicrobial peptides PGLa and magainin 2 (MAG2) from the skin of the African frog *Xenopus laevis* show a synergistic enhancement of their activity [1]. Both peptides form amphipathic α-helices upon binding to a lipid bilayer, and their orientation in membranes has been determined with high accuracy using solid state ^{2}H-, ^{15}N-, and ^{19}F-NMR.

We have previously shown that PGLa inserts upright into DMPC/DMPG bilayers in the presence of an equimolar amount of MAG2, but not on its own, not even at high concentrations [2]. This synergistic effect suggests the formation of stable heterodimeric peptide pores. Here, we show that MAG2 always stays aligned almost flat on the membrane surface, both with and without PGLa present. However, it undergoes a slight change in the orientation in the presence of PGLa, and its mobility is reduced, indicating that a PGLa-MAG2 complex is formed. The same behaviour for both PGLa and MAG2 is found in DLPC, DMPC and DPPC bilayers.

In contrast, in POPC/POPG bilayers, both peptides stay aligned flat on the membrane surface, alone as well as in 1:1 mixtures, and there seem to be no interactions between the peptides. This behaviour is also found in DMoPC (di-14:1-PC) and POPE/POPG/TOCL bilayers, indicating that the hydrophobic thickness of the membrane is not responsible for the differences. The behaviour of PGLa and MAG2 is in agreement with our previous finding that the related peptide MSI-103 always stays flat on the surface in unsaturated lipids (or more generally in lipid systems with negative spontaneous curvature), while it can tilt and insert more deeply into saturated lipid bilayers (where the spontaneous curvature is positive) [3]. Thus, the spontaneous curvature seems to be a key factor determining the membrane insertion and synergistic action of antimicrobial peptides.

References: [1] E Strandberg, P Tremouilhac, P Wadhwani, AS Ulrich (2009). *Biochim Biophys Acta* **1788**, 1667–1679. [2] P Tremouilhac, E Strandberg, P Wadhwani, AS Ulrich (2006). *J Biol Chem*, **281**, 32089–32094. [3] E Strandberg, D Tiltak, S Ehni, P Wadhwani, and AS Ulrich (2012). *Biochim Biophys Acta* **1818**, 1764–1776.