

Aggregation, Leakage and Fusion of Liposomes induced by an Antimicrobial Peptide, Hemoglobin α Chain (Hb40-61a) Fragment

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Introduction and Aims. The Hb40-61a is a synthetic amidated peptide derived from a fragment of α -chain of bovine hemoglobin (KTYFPHFDLSHGSAQVKGHGAK) isolated from cattle tick gut (*Boophilus microplus*). Antimicrobial experiments showed its potential antimicrobial activity against *Candida albicans*. Here we investigate aggregation, leakage and fusion of a membrane model induced by peptide addition. **Results.** Large unilamellar vesicles (LUVs) composed by egg-phosphatidylcholine (PC) and phosphatidylglycerol (PG) 7:3 or 9:1 w/w were prepared by injection. Peptide addition lead to LUV aggregation in both charge ratios studied and this phenomenon was ionic strength and charge dependent. As the ionic strength increased, the aggregation decreased, this effect being more pronounced with lower charged LUVs. Peptide-induced aggregation of PC/PG LUVs 9:1 and 7:3 w/w was not observed above 50 mM NaCl. Membrane fusion was estimated using an assay with PC:PG LUVs doped with PyPC. Vesicles containing PC/PG/PyPC (8.9:1:0.1 w/w) were mixed with vesicles of PC/PG (9:1w/w). After Hb40-61a addition an exponential decay of the excimer/monomer ratio occurs for different concentrations of peptide, indicating vesicles fusion. The fusion was time and salt dependent, as observed in LUVs aggregation. Leakage studies were also realized entrapping both pyrenetetrasulfonate (PTS) and the fluorescence suppressor methyl viologen. Fluorescence of the doubly doped LUVs increased with peptide concentration and decreased at higher ionic strength. **Conclusion:** Those results indicate that Hb40-61a is a fusogenic peptide and its activity is charge and salt concentration dependent.

Aknowlegments: FAPESP, CNPq

KEYWORDS: Liposome, antimicrobial peptide, leakage, fusion, Hb40-61a.