Conformational and Thermodynamic Aspects of Melittin Binding to Heparan Sulfate

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Melittin, from bee venom from *Apis Melifera*, is a 26-residue amphiphatic peptide with well known cytolytic and antimicrobial properties. Studies on the action mechanism of melittin have focused almost exclusively on the membrane perturbing properties of this peptide, investigating in detail the melittin-lipid interaction, both experimentally and theoretically. Here we report physical-chemical studies on an alternative mechanism by which melittin could interact with the cell membrane. As the outer surface of many cells is decorated with anionic (sulfated) glycosaminoglycans (GAGs), whereas melittin is positively charged, a strong interaction between two oppositely charged types of molecules can be envisaged. Indeed, isothermal titration calorimetry reveals a strong binding of melittin to a model GAG, i.e. heparan sulfate (HS). The binding constant is 2.4 x 10^5 M^{-1}, the reaction enthalpy is $\Delta H^0_{melittin}$ = -1.50 kcal/mol and the peptide-to-HS stoichiometry is ~11 (10mM Tris, 100mM NaCl, pH 7.4, 28 ºC). The reaction enthalpy becomes more negative upon an increase in temperature, and the molar heat capacity is ΔC_p^0 = -227 cal mol^{-1} K^{-1}. The large negative heat capacity change indicates that hydrophobic interactions play an important role in the binding of melittin to HS. Salt dependence studies of the binding constant confirm that electrostatic interactions between melittin and HS make an equally important energy contribution. Binding of melittin to HS is accompanied by a dramatic increase in α-helix structure.

The question arises if the interaction with sulfated glycosaminoglycans is unique for melittin or can also be observed for other peptides of this group. As an additional example we have studied interaction of HS with two other cationic peptides, magainin 2 and nisin Z, both known for their antimicrobial properties, but in contrast to melittin, not causing lysis of the eukaryotic cells. Isothermal titration calorimetry shows no binding of magainin 2 and nisin Z to HS at the temperatures of 5, 28 and 50 °C in Tris buffer

(10mM Tris, 100mM NaCl, pH 7.4). This may point to a yet unknown biological mechanism of action in case of melittin.

References:

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