diabetes. MS is a serious health problem due to its related cardiovascular disorders: hypertension and heart failure. The latter is among the major causes of death in México. The molecular mechanisms responsible for MS are unclear but could be related to anomalies in cardiac excitation-contraction coupling (E-C coupling). The cardiac Ca^{2+} channel, the Ryanodine Receptor (RyR2), is a key macromolecular complex that participates in releasing Ca^{2+} from internal stores and is centrally involved in the modulation of cardiac E-C coupling. Our aim was to examine alterations in the expression level, phosphorylation status, Ca^{2+} sensitivity and *in situ* function (Ca^{2+} sparks and Ca^{2+} transients) of RyR2 that could explain the cardiac dysfunction associated with MS.
MS was induced in our rat model by adding commercially refined sugar (30% sucrose) to their drinking water. The sucrose-fed rats became overweight with an increased accumulation of waist fat and also developed hypertension. Our [^{3}H]ryanodine binding data show that functional RyR2 are decreased in MS rat hearts with slight but insignificant changes in Ca^{2+} sensitivity. Western Blot analysis confirmed that MS did not alter the phosphorylation status of RyR2 at Serine-2809 normalized with respect to total RyR2. A significant decrease in Ca^{2+} spark frequency was found in isolated Fluo-3 loaded cardiomyocytes of MS rats. In addition Ca^{2+} transients elicited at frequencies of 0.5, 1, and 2 Hz were also impaired, suggesting a diminished Ca^{2+} cycling condition in MS cardiomyocytes.
Overall, the decreased RyR2 expression together with the impaired RyR2 function could account for the reported poor overall cardiac outcome found in this animal model of MS.

Peptide & Toxin Ion Channels

560-Pos
Effect of Dipole Modifying Agents on the Surfactin Induced Conductance of Planar Lipid Bilayers
Olga Ostroumova, Maxim Ilin, Valery Malev, Ludmila Schagina.
Institute of Cytology of RAS, St. Petersburg, Russian Federation.
We studied effects of compounds changing membrane dipole potential on membrane conductance induced by antimicrobial lipopeptide surfactin. Surfactin added on both sides of an artificial lipid bilayer from diphytanoyl phosphocholine in 1 M KCl (pH 6.5) produces an increase of the membrane conductance as a result of ion channel formation. Increasing a membrane dipole potential adding RH 421 to the bilayer bathing solution (10 μM , both sides) leads to ~40 times increase of a steady-state conductance. At the same time, addition of phloretin (20 μM), known to decrease the dipole potential, results in decrease of the surfactin-induced conductance by ~30 times. We note, that the effects of dipole modifiers on the surfactin-induced membrane conductance are clearly opposite to the effects observed with the same modifiers in case of syringomycin E-induced conductance of lipid bilayers [Ostroumova et al., Langmuir, 2008]. As we suggested earlier, the influence of dipole modifiers on syringomycin activity may be related to a promotion/retardation of a movement of positively charged syringomycin molecules in the direction of membrane hydrocarbon core. In contrast to syringomycin, surfactin is negatively charged. Hence, one can expect an inversion of the effect of dipole modifiers in case of surfactin. The obtained results are in agreement with the model proposed in [Ostroumova et al., Langmuir, 2008].
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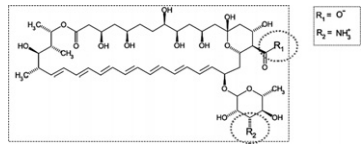
561-Pos
Effect of Antibacterial Peptide Indolicidin on the Membrane Permeability: Carrier Mechanism Versus Pore Formation
Tatyana I. Rokitskaya[1], Nikolay I. Kolodkin[2], Yuri N. Antonenko[1].
[1]Belozersky Institute of Physico-Chemical Biology, Moscow State University, Moscow, Russian Federation, [2]State Research Institute of Highly Pure Biopreparations, St Petersburg, Russian Federation.
It is generally accepted that the predominant mechanism of action of antimicrobial peptides is the permeabilizaiton of bacterial membranes via formation of aqueous pores. It has been shown in the present work that the main mechanism of carboxyfluorescein (CF) leakage from lipid vesicles induced by antimicrobial peptide indolicidin is not pore formation but rather translocation across the membrane of the complexes of the dye and the peptide, i.e. indolicidin functions as a carrier of organic anions. This conclusion was made after observation of strong inhibition of CF leakage by other organic anions (such as fatty acids) and also inability of indolicidin to induce leakage of glucose and positively-charged doxorubicin. Besides, formation of complexes of indolicidin with pyrenebutanic acid was directly observed by fluorescent assay. The mode of action proposed here for indolicidin can be related to that previously postulated for oligoarginine derivatives which are able to transport organic anions across liposomal and bulk phase membranes [Sakai N. et al., ChemBioChem. 2005, 6:114-122]. The newly identified mechanism may be involved in bactericidal action of indolicidin either directly or indirectly through induction of leakage of important anionic metabolites leading to regulatory disfunction.

562-Pos
Molecular Action Mechanism of Amphotericin B and Structural Analogs on Biological Membranes
Mauricio Carrillo-Tripp[1], Alex H. de Vries[2], Rogelio Hernández[3], Cristina Vargas[1], Humberto Saint-Martin[3], Ivan Ortega-Blake[3].
[1]Cinvestav, Merida, Mexico, [2]Biomolecular Sciences and Biotechnology Institute, Groningen, Netherlands, [3]UNAM, Cuernavaca, Mexico.
Amphotericin B is a polyene antifungal drug, often used intravenously for systemic fungal infections. It is believed that AmB associates with ergosterol, the main component of fungal cell membranes, forming a transmembrane channel that leads to K+ leakage and fungal cell death. United atom Molecular Dynamics simulations where used to study the mechanism of action of AmB, and other structural analogs, on a POPC and ergosterol bilayer (3:1), varying toxins concentration. It is shown that toxins aggregate first in solution before adsorbing into the membrane, both at low and high concentrations. Electrostatic properties of AmB play an important role in toxicity and sterol selectivity in comparison with AmB's structural analogs.



563-Pos
Divalent Cations Regulate Pore Formation of Synthetic, Naturally Occurring Alamethicin and Selected Analogs
Mascia Benedusi, Alberto Milani, Marco Aquila, **Giorgio Rispoli**.
Università di Ferrara, FERRARA, Italy.
The biophysical characteristics of synthetic, naturally occurring peptides forming membrane-spanning channels were investigated by using isolated rod outer segments (OS) of frog, recorded in whole-cell configuration. The peptides were applied to (and removed from) the OS in ~50 ms with a computer-controlled microperfusion system. Once blocking the main OS endogenous conductance (the cGMP channels) with saturating light, the OS membrane resistance was mostly >1 GOhm. In symmetric K or Na, 1 μM synthetic alamethicin F50/5 produced a current after ~0.21 s from the solution exchange (called *Delay*), that activated monoexponentially (time constant τ_a~0.26 s) to a maximal amplitude (I_{max}) of ~700 pA. Peptide removal caused the current to return to 0, with a non-measurable *Delay*, and again monoexponentially (time constant τ_d~0.31 s), showing the full reversibility of the permeabilization process. I_{max}, *Delay*, τ_a, and τ_d of current produced by 1 μM of [L-Glu(OMe)^{7,18,19}] (where the Gln 7,18,19 were substituted with side-chain esterified Glu residues) were respectively similar, ~8-fold, ~16-fold, and ~6-fold larger than those of alamethicin F50/5. For both peptides, the current-to-voltage characteristics (obtained with voltage ramps) showed a strong inward rectification at early times of application; current was carried equally well by monovalent and divalent cations. However, activation kinetics accelerated more than 100-fold if external Na or K was substituted with an equiosmolar amount of Ca in the case of F50/5, but up to 10-fold in the case of [L-Glu(OMe)^{7,18,19}]; *Delay* and τ_d were not significantly affected by divalent cations. Similar results were preliminarily obtained in the presence of Mg or Mn, indicating that the effect of divalent cations was not due to a change in the surface charge density of plasma membrane, but to an increase of probability of pore formation.