presented. The impact of the AMP maculatin 1.1 (Mac1) on bacteria was monitored by ^{31}P while structural details on the peptide were obtained using dynamic nuclear polarization (DNP) enhanced ^{13}C and ^{15}N solid-state NMR experiments. Under AMP stress, a significant change in DNA packing in *E. coli* and *S. aureus* was observed. Mac1 also modulated the lipid dynamics of the bacterial membranes. Finally, a novel strategy to perform in-cell DNP NMR experiments was established by using spin-labelled peptides; and $\{^{15}N\}^{13}C$ REDOR measurements have been performed to measure the distance between several pairs of ^{13}C=O and ^{15}NH within the Mac1 amino acid sequence, which indicate that the peptide adopts a helical structure in bacteria.

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Solid-State NMR of Intact Bacteria Reveals the Effect of Stress and Antimicrobial Agents

Zeineb Bouhlel[1], Dror E. Warschawski[2], Alexandre A. Arnold[1], Karine Lemarchand[3], Réjean Tremblay[3], **Isabelle Marcotte**[1].
[1]Chemistry, Universite du Quebec a Montreal, Montreal, QC, Canada, [2]Institut de Biologie Physico-Chimique, CNRS-Universite Paris Diderot, Paris, France, [3]Institut des Sciences de la Mer, Universite du Quebec a Rimouski, Rimouski, QC, Canada.

The worldwide problem of bacterial resistance requires the development of novel antibiotics against bacterial pathogens. It is thus essential to determine the action mechanism of these drugs, and in particular, to understand their interaction with the bacterial cell wall - the first barrier crossed or, in some cases, targeted. ^{2}H solid-state (SS) NMR is a valuable tool to probe, at a molecular level, the effect of antimicrobial agents on the organization and dynamics of membrane phospholipids. These studies are usually performed on model membranes, but ''in-cell'' SS-NMR is advantageous as it takes into account the complexity of the cell wall in the interaction. We developed for the first time a protocol to deuterate the membrane phospholipids of *Vibrio splendidus* - a Gram(-) marine bacterium. To do so, we used deuterated palmitic acid with oleic acid to better preserve the natural membrane saturated/unsaturated lipid ratio. We characterized the membrane fluidity *in vivo* during different growth stages by ^{2}H SS-NMR using magic-angle spinning (MAS) and spectral moment analysis. Our results show greater membrane fluidity during the stationary growth phase compared to the exponential phase under labelling conditions. The difference in membrane fluidity is related to a change in fatty acid chain composition, indicating that bacteria adapt to cope with the ''stress'' induced by the labelling conditions. Then *in vivo* ^{2}H SS-NMR was used to study the interaction of a promising antimicrobial pigment called ''marennine'' on *V. splendidus* membranes. Our results suggest that marennine would act through a stiffening mechanism that could involve interaction with lipopolysaccharides in the outer membrane. Altogether, our work shows that ^{2}H SS-NMR is a useful tool to reveal the effect of stress and interactions on the membrane of intact microorganisms.

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The Ebola Virus Δ-Peptides are Enterotoxic Viroporins *In Vivo* and Potentially Druggable Targets

Shantanu Guha[1], Lilia Melnik[2], Jenisha Ghimire[1], Allison Smither[2], Eric Wu[1], Leisheng Sun[1], Nathan Ungerleider[3], Erik Flemington[3], Robert F. Garry[2], William C. Wimley[1].
[1]Dept of Biochemistry & Molecular Biology, Tulane University School of Medicine, New Orleans, LA, USA, [2]Dept of Microbiology & Immunology, Tulane University School of Medicine, New Orleans, LA, USA, [3]Dept of Pathology, Tulane University School of Medicine, New Orleans, LA, USA.

Ebola virus (EBOV) is a causative agent of viral hemorrhagic fever and remains an ongoing threat to global public health due to large outbreaks of the virus. EBOV and many other filoviruses generate a peptide cleavage product from their glycoprotein genes during active pathogenesis called the delta peptide, but the function of this peptide has been unknown. Here, we describe the activity of the EBOV delta peptide in the context of the small intestine. *In vivo,* the delta peptide causes severe fluid accumulation and diarrheal syndrome within a closed ileal loop in BALB/C mice. This activity is dose-dependent and peaks 9 to 12 hours after introduction of the peptide to the closed intestinal loop. Various truncated delta peptides containing the conserved C-terminus have similar viroporin activity. The EBOV delta peptide may function through the formation of multimeric ion channels in cell membranes, causing ion dysregulation, a hallmark of diarrheal syndrome and gastrointestinal distress. These findings demonstrate that the EBOV delta peptide is an enterotoxic viroporin and may be categorized as a novel virulence factor. Subsequently, we have engineered an antigen by conjugating a delta peptide variant (E23ox) to a carrier protein and immunized rabbits to generate custom high-affinity binding antibodies to neutralize viroporin activity. Our preliminary data suggests that the Ebola virus delta peptides are druggable targets and a potentially worthwhile avenue for novel therapeutic development.

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Munc13 Clusters Capture Vesicles to Lipid Bilayer Membrane

Feng Li[1], R. Venkat Kalyana Sundaram[2], Jeff Coleman[1], Shyam S. Krishnakumar[1], Frederic Pincet[2], James Rothman[1].
[1]Cell Biology, Yale University, New Haven, CT, USA, [2]Yale University, New Haven, CT, USA.

Munc13 is a large protein and has complex functions in synaptic vesicle fusion. Recent studies suggested that the release sites in the active zones of neurons consist of Munc13 nano-assemblies. However, molecular mechanism of formation of Munc13 cluster and its function are unclear. Here we reconstituted Munc13 on lipid bilayer membrane and found Munc13 clusters of various sizes were formed. We used TIRF microscope to perform step bleaching of the clusters and were able to determine the copy numbers of Munc13 in the clusters. We found that Munc13 clusters, rather than monomeric Munc13 molecules, are able to capture vesicles from solution to membrane. Moreover, a threshold of the copy number in clusters exists. When the copy number of Munc13 is larger than the threshold, the clusters are capable of capturing vesicles; if smaller, the clusters are not able to capture vesicles. DAG lipid or phorbol ester is required for Munc13 to form clusters larger than the threshold, and therefore, is critical to Munc13's capability of capturing vesicles.

In the absence of PIP2, Munc13 still form clusters, with similar size distribution, but clusters' capability of capturing vesicles is largely decreased. It is likely that interaction between PIP2 and Munc13 facilitates Munc13 to adopt the proper orientation on membrane for vesicle capture.

A Munc13 mutant, in which the C2C domain is deleted, still forms clusters on membrane. But the efficiency of vesicle capture by such clusters is significantly reduced.

As the interaction range of Munc13 is larger than Synaptotagmin and SNAREs, our results suggest that Munc13 clusters have an essential function to initiate chaperoning synaptic vesicles to the active release sites in synapse. Our results also shed new light on the structure and dynamics of Munc13 clusters.

Platform: Cardiac, Smooth, and Skeletal Muscle Electrophysiology and Regulation I

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Quantitative Cross-Species Prediction of β-Adrenergic Response in Ventricular Myocytes

Stefano Morotti, Haibo Ni, Lianguo Wang, Alex Fogli Iseppe, Donald M. Bers, Andrew G. Edwards, Crystal M. Ripplinger, Eleonora Grandi.
Department of Pharmacology, University of California Davis, Davis, CA, USA.

Despite the large body of research on neural control of the heart, our quantitative understanding of the role of β-adrenergic receptor (β-AR) stimulation in human cardiac electrophysiology and arrhythmogenesis remains poor. One possible reason is in the fact that our knowledge is mostly based on experimental animal models (e.g., mouse and rabbit) characterized by quite different cellular electrophysiology. Indeed, we have shown that well conserved mammalian responses to β-AR stimulation (fight or flight) are mediated by different sub-cellular processes in mouse vs. rabbit. Here, we aim to quantitatively assess these inter-species differences, and to construct a suite of tools for mapping physiological responses across species. We simulated our mathematical models of excitation-contraction coupling in mouse, rabbit and human ventricular myocytes, integrating detailed formulation of β-AR signaling, to generate populations of models by random parameter variations. Population-level analysis of action potential (AP) and calcium transient (CaT) properties, at both baseline and upon β-AR activation, allowed quantifying inter-species differences in the sensitivity of these features to changes in model parameters. We then utilized statistical regression models to develop quantitative predictors of human response from different subsets of simulated AP and CaT data (mimicking variable composition of experimental datasets). Predictors built upon calibrated AP and CaT signals can well reconstruct human response from mouse or rabbit measurements. However, predictors devoid of calibrated (absolute) measures performed more poorly. In the absence of calibrated data, prediction of human response could be improved by combining both mouse and rabbit data. Further refinement and validation of these quantitative models will provide new tools to understand inter-species difference in the response to sympathetic stimulation and might aid future therapeutic strategies based on neuromodulation.