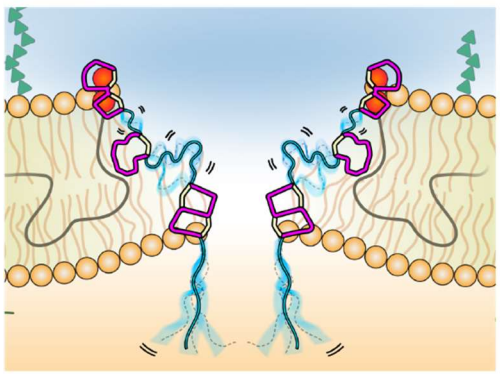
Biophysics of Peptide Antibiotics and Viral Channels in Lipid Membranes, with Solid-State Nuclear Magnetic Resonance

Understanding the interplay between lipids and membrane proteins is crucial for new advances in molecular medicine, such as identifying the molecular basis of disease, developing new antibiotics and elucidating drug targets. Despite its critical importance, the tools for studying membranes systems at the molecular level are limited. Solid state Nuclear Magnetic Resonance (ssNMR) stands-out as a non-invasive spectroscopic technique capable of examining membrane systems in their natural state, providing insights into their molecular structure, dynamics and interactions.

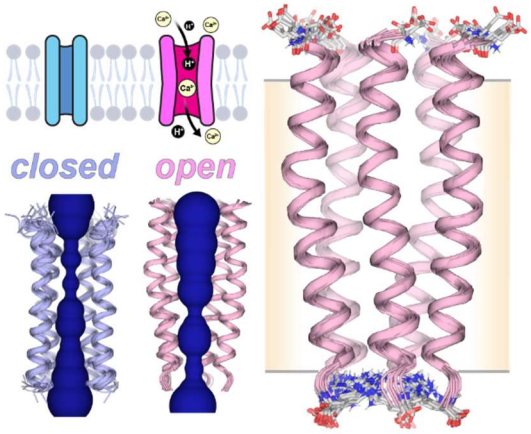
We developed an innovative ssNMR methodology to study the structure, binding and dynamics of peptide antibiotics directly within cell membranes, thereby uncovering their medically significant modes of action. For example, we show that the native binding mode of the pore-forming antibiotic Nisin strongly deviates from previously published structures. Notably, Nisin revealed structural motifs that adapt its molecular structure to the specific properties of the target cell membrane. Additionally, we elucidate how Teixobactin, a highly promising clinical antibiotic, targets the essential bacterial Lipid II. Teixobactin binds Lipid II at the membrane surface, initiating the formation of amyloid-like fibril structures that disrupt the bacterial membrane, leading to cell death. This is a novel antimicrobial mode of action that represents a paradigm shift in our understanding of lipid-targeting antibiotics.

ssNMR is also invaluable for analyzing viral channels, as I demonstrate in our study of the Envelope (E) Protein of SARS-CoV-2. The E Protein forms a cation channel that disrupts the pH and calcium homeostasis in infected cells, causing severe viral-induced inflammation. In mice, inhibiting the E channel activity attenuates the pathogenic effects of the virus, making it an attractive antiviral target for treating COVID-19. Using ssNMR, we determined the structure and gating mechanism of the E protein in lipid membranes. We found that the E channel forms a pentameric α-helical barrel that adopt a cylindrical structure in the closed state, and a conical structure in the open state. Our findings further suggest that the channel gating is governed by polar interactions at the pore-openings and an aromatic network at the channel core. This detailed structural insight into the E's ion conduction paved the way for the rational design of new drugs for treating COVID-19.

Native-State Structure of Antibiotics



SARS-CoV-2 E viroporin



About the speaker

João Medeiros Silva is original from Porto Formoso, São Miguel. He studied Biochemistry at NOVA Lisbon University and was a research fellow at the Health Sciences Research Center in Covilhã. From 2016-2020 he obtained his PhD with highest distinction from Utrecht University in the Netherlands.

Dr. Silva's core expertise lies in Nuclear Magnetic Resonance (NMR) spectroscopy. He has developed NMR techniques to describe the functioning and biophysical properties of membrane proteins and lipids down to the atomic level. As result, he has elucidated the ion transport of potassium channels and the mechanism of action of some of the most clinically promising antibiotics. His work has received several distinctions, including awards from the Portuguese Chemical Society, and the Netherlands' Societies for Biochemistry, Biophysics and Magnetic Resonance.

Since 2020, he is a postdoctoral researcher at the Massachusetts Institute of Technology. His current research focuses on investigating the structure and mode of function of ion channels from SARS-CoV-2, and elucidating how these proteins induce acute inflammation in infected cells. These findings have facilitated the development of new drugs for treating COVID-19 patients. His research is supported by the Netherlands Organization for Health Research and the European Molecular Biology Organization (EMBO).

Representative publications:

1- Medeiros-Silva, J.; Dregni, A.J.; Somberg, N.H.; Duan, P.; Hong, M.; **Atomic structure of the open SARS-CoV-2 E viroporin.** (2023) *Science Advances*, 9, 41

2- Shukla, R*; Medeiros-Silva, J.*; Parmar, A.; Vermeulen, B.; Das, S.; Lucini Paioni, A.; Jekhmane, S.; Lorent, J.; Bonvin, A.M.J.J.; Baldus, M.; Lelli, M.; Verldhuizen, E.; Breukink, E.; Weingarth, M.; **Mode of action of teixobactins in cellular membranes.** (2020) *Nature Communications*, 11, 2848.

3- Medeiros-Silva, J.; Jekhmane, S.; Lucini Paioni, A.; Gawarecka, K.; Baldus, M.; Swiezewska, E.; Breukink, E.; Weingarth, M.; **High-resolution NMR studies of antibiotics in cell membranes.** (2018) Nature Communications, 9, 3963.