Experimental report

09/04/2020

Proposal: **9-13-828** **Council:** 4/2019

Title: EINS study of a model protomembrane sub-ns dynamics under high temperature and pressure

Research area: Soft condensed matter

This proposal is a new proposal

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Samples: decanol
decanoic acid
squalane
eicosane

Instrument	Requested days	Allocated days	From	To
IN13	11	11	07/02/2020	18/02/2020

Abstract:

A novel membrane architecture is proposed as a model for the first forms of life.
The model consists on a bilayer made of short single chain amphiphiles with an intercalating alkane in the membrane midplane. Such model, which takes into account the limitations in molecular complexity in the early Earth environment, could also explain how the first protocells survived to the extreme environmental conditions. The hydrocarbon molecules, in fact, should allow changing the physico-chemical properties of the membrane, allowing to keep the biological functionality at higher temperature and pressure values.
SANS studies gave positive results about the model structural characterization, showing that the alkanes intercalate in the membrane and affect its structure.
We propose here to focus on the amphiphilic hydrogen dynamics using IN13, to study how this is affected by the alkane inserption. The experiment will allow a more general overview by exploring the combined effects of temperature and pressure on the model membrane.

Exp. Report 9-13-828

EINS study of a model protomembrane sub-ns dynamics under high temperature and pressure

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One of the central question in the emergence of life is the invention of the cell membrane. In contemporary cells, the membrane is essential since it hosts the energy harvesting machinery driven by the proton motive force. If, as suggested by most origin of life scenarios, life originated in deep-sea hydrothermal vent [1], early cells had to cope with a very challenging environment (strong gradient in temperature and chemical composition) as well as high hydrostatic pressure. These models also suppose that membranes were build out of major amphiphiles of the early Earth, e.g. simple organic acids or alcohols readily available [2], which would create membranes of much lesser stability than modern membrane lipids. To circumvent this apparent contradiction, we have proposed that early membranes would have been based of simple, single chain amphiphiles (i.e. decanoic acid/decanol), with an alkane (i.e. eicosane, squalane) inserted in its midplane. This architecture was recently proposed to explain the stability of the lipid bilayer in modern polyextremophiles [3].

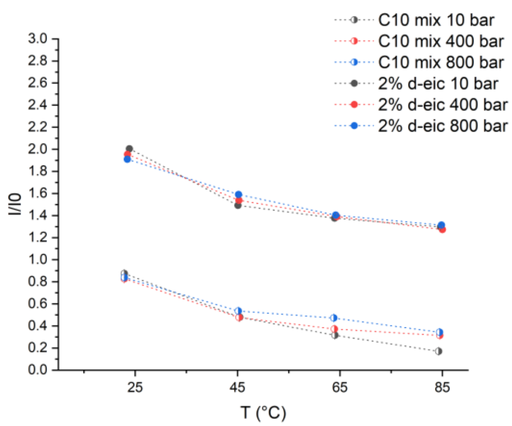
Indeed, by populating the membrane midplane the alkanes will shift the functional domain of the membrane towards higher pressure and temperature [4]. If the functional predictions are demonstrated for simpler lipid molecules, these effects would highlight a possible strategy to explain how the first living systems maintained and protected the biological functions of its boundaries at extreme T and p prebiotic conditions.

Previous studies were made using Small Angle Neutron Scattering (SANS) to investigate the effects of the presence of alkane on the membrane form factor using Uni- and Multilamellar Vesicles (ULVs - MLVs). The obtained results (DOI:10.5291/ILL-DATA.9-13-788 and related Exp. Report) allowed us to prove that the alkanes intercalate in the membrane and affect its structure as well as its stability with increasing temperature. However, to account for the above-mentioned early planet's conditions, a more complete study needs to be done by changing pressure and temperature together, so that the behavior of the model vesicles is investigated in the whole p-T region of interest: $20 \leq T \leq 80$ °C and $1 \leq p \leq 800$ bar.

Elastic Incoherent Neutron Scattering (EINS) can be used for this purpose, as a tool to study the effects of the hydrocarbon presence inside the membrane on the averaged dynamics of its components (the amphiphilic chains) as a function of T and p. The following samples were then probed:

- C10 mix (Decanoic Acid + Decanol 1:1) - vesicles without alkanes
- C10 mix + perdeuterated eicosane (2% d-eic)– addition of a linear alkane, which is invisible to neutrons
- Capric acid – vesicles without alcohol and alkanes (the HP cell was broken at higher temperature for this last sample, so we are not showing the incomplete data set of this sample)

We calculated the normalized intensities summed over all scattering angles of the samples as function of T and p (see figure 1). All curves are almost superposed as function of pressure, but the intensities decrease as expected as function of temperature. One can see a small divergent behavior at high temerpatures for the C10 mix.



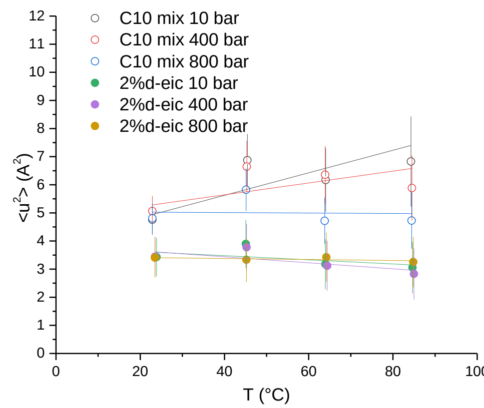
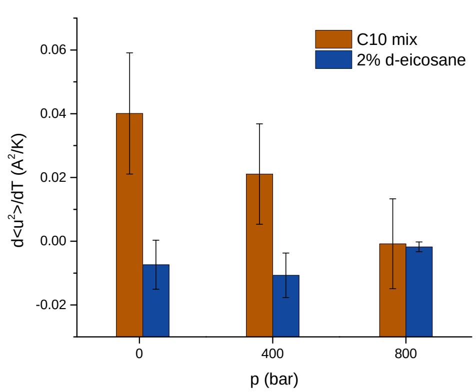


Figure 1: Normalised summed intensities of both samples as function of T and p.

Figure 2: Mean square displacements of both samples as function of T and p.

This is further confirmed when plotting the the atomic mean square displacements $<u^2>$ as function of T and p (see figure 2). In absence of the alkane in the C10 mix sample, the $<u^2>$ are increasing with temperature at 10 and 400 bar, but at the highest pressure of 800 bar the sample become insensitive to temperature increase. In contrast, eicosane has the clear effect to stiffen the vesicles and to make the sample insensitive against pressure and temperature changes showing its potential to always maintain the vesicles in a constant state even under extreme p and T conditions.



We summarize these findings in figure 3 where we show the slopes of the mean square displacements against T of the two samples for the three pressure values. As shown, the slope varies greatly for the C10 mix at the 3 pressures, while the addition of eicosane makes the system almost insensitive to pressure variations. This demonstrates that linear alkanes could have served as a valuable tool to suppress the protomembrane sensitivity towards extreme temperature and pressures, and therefore allow their existence in the very variable physico-chemical conditions of deep sea hydrothermal vents at the origins of life.

Figure 3: Slopes of $d<u^2>/dT$ for both samples as function of p.

Literature

[1] M.S. Dodd; D. Papineau ; T. Grenne ; J.F. Slack; M. Rittner; F. Pirajno; J. O'Neil; C.T.S. Little, Nature 2017 543, 60–64.
[2] P.M. Monnard; D.W. Deamer, Met. Enzym. 2003, 372:133-51.
[3] A. Cario; V. Grossi; P. Schaeffer; P.M. Oger, Front. Microb. 2015, 6:1152.
[4] M. Salvador Castell et al., BBA Biomembranes 1862 (2020) 183130.