Experimental report

15/01/2022

Proposal: 9-13-935 **Council:** 4/2020

Title: Neutron Reflection Study of the interaction of DPPC/DPPG monolayer with acyl-L-carnitine in different pHby Langmuir Trough

Research area: Physics

This proposal is a new proposal

Main proposer: Jian Ren LU

Experimental team: Armando MAESTRO

Local contacts: Armando MAESTRO

Samples: DPPC/chain deuterated DPPC
DPPG/chain deuterated DPPG
C16Lcarnitine(C23H45NO4)/chain deuterated C16LC
C12Lcarnitine(C19H37NO4)/chain deuterated C12LC
C14Lcarnitine(C21H41NO4)/chain deuterated C14LC

Instrument	Requested days	Allocated days	From	To
FIGARO	4	3	12/03/2021	15/03/2021

Abstract:

In the past decade, there is a rise in the disease caused by bacterial infections. As a result, there is a high demand for the antibacterial agents which can kill bacteria effectively, especially in some extreme environment. From the pre-work, we measured the CMC of C12-L-carnitine, C14-L-carnitine and C16-L-carnitine, and their MIC to several kinds of bacteria e.g. E.coli. We found that in pH 7, acyl-L-carnitines have no effect to E.coli. But when the in pH 5, acyl-L-carnitines can kill E.coli in a low concentration. This means acyl-L-carnitines have a pH-responsive antibacterial function. However, there is no direct structural evidence to study how acyl-L-carnitines interact with bacteria. Neutron reflection will examine how acyl-L-carnitine effect the membrane model of bacteria. The success of this proposal will provide constructive suggestions for further carnitine design.

Experimental report

03/2020

Proposal: 9-13-935

Council:

Title: Neutron Reflection Study of the interaction of DPPC/DPPG monolayer with acyl-L-carnitine in different pHby Langmuir Trough

Research area: Biosurfactants,

Main proposer: **Jian Ren LU**

Experimental team: Huayang LIU

Zongyi LI

Ke FA

Local contacts: Armando MAESTRO

Samples: Acyl-L-carnitines

Instrument	Requested days	Allocated days	From	To
FIGARO	3	3	12/03/2021	15/03/2021

Abstract:

Acyl L-Carnitines are a group of novel surfactants with attractive physical and biological properties associated with their head group charge features. This work proposes to use neutron reflection as the unique technique to study how acyl-L-carnitine adsorb on the silica/water interface. We request 3 days of FIGARO beam time to complete the proposed measurements

Report of experiment – FIGARO 9-13-935

PhD Project: Study the fundamental properties of acyl-L-carnitines

Huayang LIU

Because of the COVID-19, we can not go to the ILL to do the experiment by ourselves and we have to post our samples to ILL. So we could not finish the SiO_2/water adsorption in 9-10-1621. The purpose of this experiment is continue to study the adsorption of acyl-L-carnitines in silica/water under different concentrations, pH and ionic strength. We use the FIGARO to measure the different contrasts and fit them to have the structural details of adsorbed acyl-L-carnitines layers.

Materials / samples:

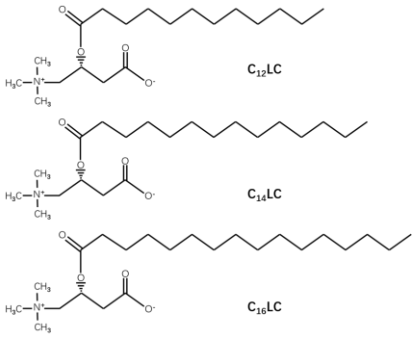

Figure 1 Chemical structure of acyl-L-carnitines

The structure of acyl-L-carnitines are shown in Figure 1. The surfactants have been purified by crystallization method to remove acid and salt, so that the samples have very high purity. The sample powder is dissolved in water with different concentrations, and the pH is adjusted with HCL solution. Then NaCl solution is added to control the ionic strength. Besides, three contrasts are measured in this experiment: h-acyl-L-carnitines are dissolved in D_2O, d-acyl-L-carnitines are dissolved in null reflection water(NRW) and D_2O. The ready samples are poured into the Teflon troughs and measured at 25℃.

Experiment and results

Due to COVID-19, this experiment continued last FIGARO experiment (adsorption at SiO_2/water interface). Firstly, we measured $C_{12}LC$ at pH 7 with low and high ionic strength (1 and 151 mM) with 3 contrasts as shown in Figure 2 (a) and (b). The best fit parameters showed that the thickness and adsorbed amount has little difference between different ionic strength at pH 7. So the ionic strength has no effect on the adsorption of $C_{12}LC$ when it is zwitterionic form. When the pH decrease to pH 3 the thickness and adsorbed amount decreased as shown in Figure 2 (c). However, the ionic strength can screen the repulsive interaction between cationic $C_{12}LC$ molecules as shown in Figure 2 (d). Besides, $C_{14}LC$ and $C_{16}LC$ were measured in the same way, the results showed that they have similar behavior as $C_{12}LC$, but the thickness of adsorbed layer increase with the acyl chain length.

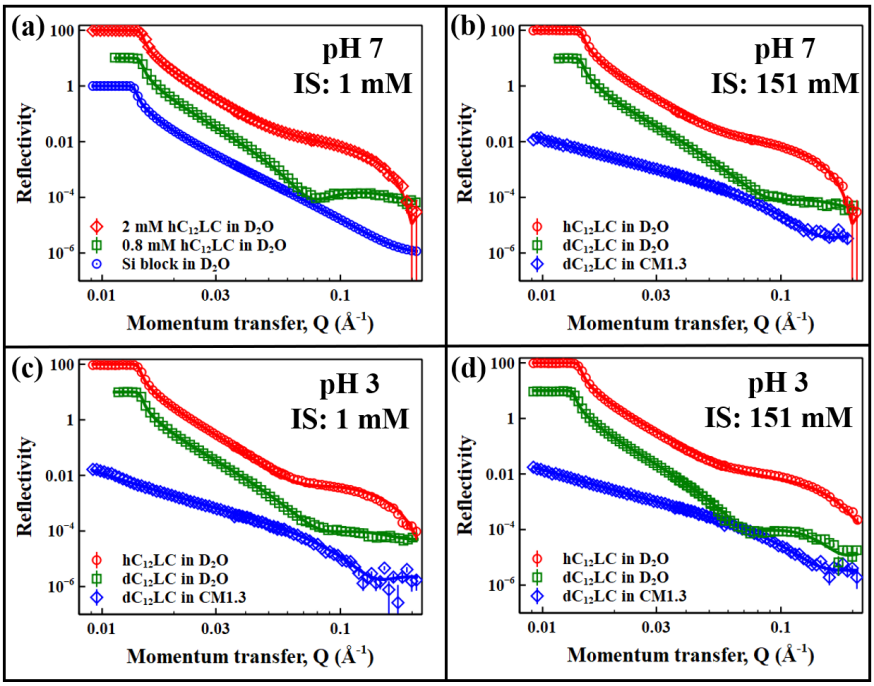

Figure 2 (a) Neutron reflection profiles of $C_{12}LC$ at pH7 and different concentrations (0, 0.8 and 2 mM). (b) Neutron reflection profiles of $C_{12}LC$ for 3 contrasts at pH7 with ionic strength of 151 mM. (c) Neutron reflection profiles of $C_{12}LC$ for 3 contrasts at pH3 with ionic strength of 1 mM. (d) Neutron reflection profiles of $C_{12}LC$ for 3 contrasts at pH3 with ionic strength of 151 mM.

Segments	Scattering length density/ρ($Å^{-2}\times10^{-6}$)
Protonanted tail ($C_{11}H_{23}$)	-0.40
Deuterated tail ($C_{11}D_{23}$, 98%D)	7.01
Protonanted tail ($C_{13}H_{27}$)	-0.39
Deuterated tail ($C_{13}D_{27}$, 98%D)	7.03
Protonanted tail ($C_{15}H_{31}$)	-0.37
Deuterated tail ($C_{15}D_{31}$, 98%D)	7.00
Head(hL-carnitine with -C=O)	1.31
Null reflection water(NRW)	0
D_2O	6.35

Table 1 The SLD of different segments used in fitting NR data

Conclusion

The adsorption amount and thickness of acyl-L-carnitines at the interface of SiO_2/water decrease as the pH decreases.

The repulsive interaction between cationic acyl-L-carnitine molecules can be screened by high ionic strength.