Experimental report

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Title: Internal layer structure a novel lipid/graphene biomembrane superstructure

Research area: Soft condensed matter

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Samples: 1,2-Dipalmitoyl-sn-Glycero-3-Phosphocholine (DPPC)
monolayer CVD graphene

Instrument	Requested days	Allocated days	From	To
D17	3	3	09/03/2018	12/03/2018

Abstract:

Understanding the interactions between lipids and graphene promises to be effective towards measuring experimentally biochemical phenomena within lipid monolayers and bilayers. Our initial results show that the deposition of graphene on a lipid monolayer results in a more ordered and extended monolayer than regions without graphene. The lipid molecules change their conformation in presence of graphene on top, perhaps suggesting that the lipids follow the natural wavy conformation of graphene. Neutron reflectometry will help us understanding the molecular organization within the layered lipid/graphene and lipid/graphene/lipid hybrid monomolecular films and get further insights on how such graphene-stabilized lipid monolayers could be lifted from their supports without disrupting the lipid-lipid and graphene-lipid interactions.

Experimental Report-Experiment n° 9-13-734

Internal layer structure a novel lipid/graphene biomembrane superstructure

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1 Context

Graphene is typically supported - sometimes sandwiched - with other two-dimensional (2D) materials to promote higher mobility, to ensure the reproducibility in electrical performances and to prevent environmental contamination [1, 2]. Frequently composed of inorganic, hard and crystalline materials, the so called van der Waals heterostructures have emerged as a route to design new and remarkably complex layer-by-layer films of 2D materials, including graphene [3]. One challenge associated with 2D materials as supporting and sandwiching layers is their limited chemical diversity, functions, and inherent inorganic nature. The possibility of combining graphene with soft, dynamic and molecular self-assembled monolayers is therefore of high interest as an organic alternative to inorganic 2D materials and could provide a versatile platform for applications, such as biosensors, drug delivery systems or cellular devices [4].

Lipids - main constituents of cell membranes - can self-assemble and form stable quasi two dimensional fluid membranes [5]. Lipids can spread on graphene [6], however little is known on the formation, stability and molecular structure of phospholipids molecules surrounding graphene [6, 7]. Mainly, studies focused on graphene oxide (GO), as both lipid vesicles and GO form stable suspensions in aqueous environments [8, 9]. GO is an easily accessible form of graphene, suitable to study the influence of oxidation states on the chemical characteristics of GO-lipid assemblies, at the cost of lower electron mobility, higher chemical reactivity, oxygen doping, and surface/edge inhomogeneities. Being negatively charged, GO has a particular affinity with positively charged lipid head groups [10], highlighting the importance of electrostatic interactions in the assembly process [11]. Pristine graphene, however, does not contain charges on the basal plane therefore minimizing electrostatic interactions and favoring hydrophobic interactions between lipid tails and graphene at the interface [12].

2 Experiments done

We performed experiments on the D17 reflectometer, in TOF mode, with wavelength range from 2 to 20 Åand two incident angles. Samples were prepared at ILL using the Soft Matter Lab facilities for the Langmuir-Blodgett and Langmuir-Schaefer techniques. A CVD graphene layer was transferred above the lipid film by bringing it into contact with graphene floating on ammonium persulfate solution (APS) as described in [13]. We have done two types of experiments : (i) graphene deposited on mono and trilayer at solid/air interface; (ii) graphene layer sandwiched between two lipid monolayer at liquid/air interface.

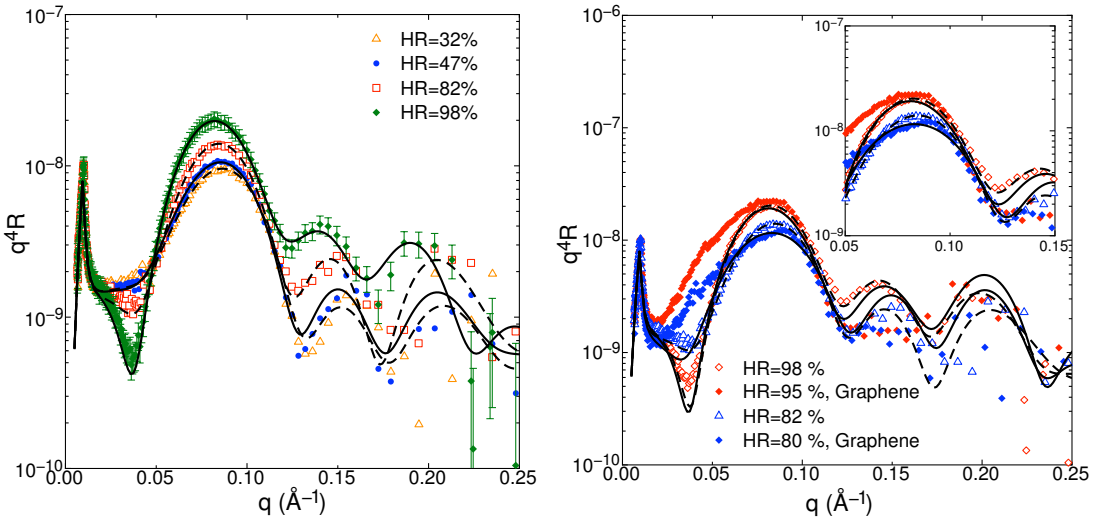
Using hydrogenated lipids we investigated different samples:

1. 2 silicon blocks characterized in 3 contrasts (H_2O, D_2O and SLD=4);
2. 1 DPPC monolayer before and after graphene transfer;

3. 3 DPPC trilayers before and after graphene transfer;
4. 1 DSPC monolayer before and after graphene transfer;
5. 1 DSPC trilayer at 3 humidity rates (HR=10, 40 and 90 %) before and after graphene transfer;
6. 1 DSPC lipid bilayer/graphene at T=20°C (gel phase and 50°C fluid phase) in 3 contrasts.

3 Results

3.1 Hybrid graphene-trilayer system



(a) (b)

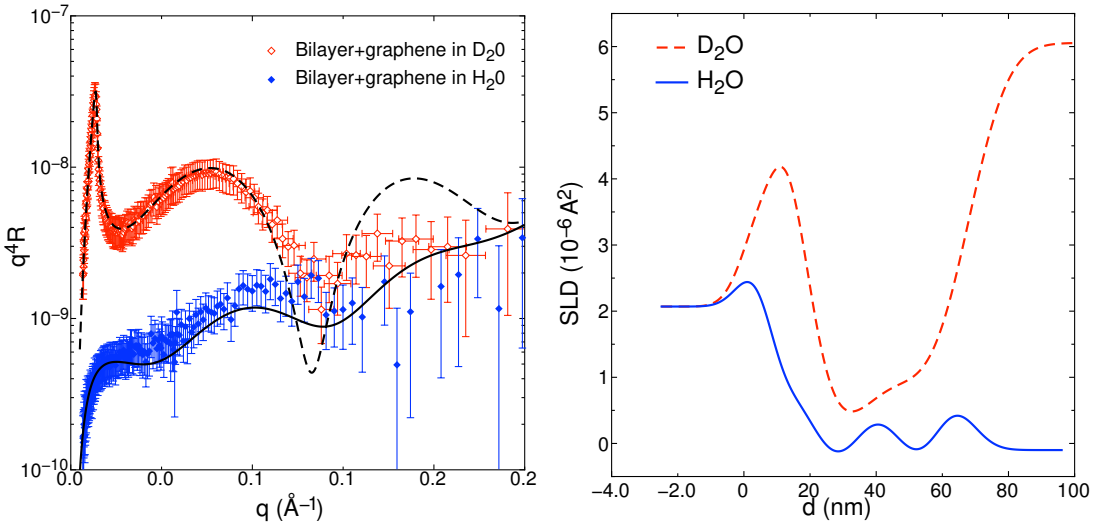
Figure 1: (a) DSPC trilayer at 25°C (gel phase) and at different humidity rate. (b) Same DSPC hybrid trilayer with a graphene layer on top. Lines corresponds to the best fit.

We have firstly checked the bare silicon block. Surprisingly, one block was presenting a large silicon oxide layer (3 nm compare to the usual 1 nm thickness) although the blocks were freshly polished. Probably because of this, we encountered great difficulties in forming good quality DPPC trilayers and only 1 of the 3 samples was actually exploitable. On contrary, we obtained an almost perfect sample of DSPC trilayer, which allowed to probe in a very reproducible way the swelling effect driven by the humidity rate, consisting in an increase of the water thickness between monolayer and the bilayer (see Fig. 1.a).

The transfer of graphene on trilayer (DPPC and DSPC) was made using a method developped in the group of Leiden University [13]. It was very difficult to reproduce graphene deposition on large samples, and we clearly need to improve our deposition techniques. Nevertheless, it is clear that the graphene layer transfer was successful, even if not homogeneous. The reflectivity data exhibit a large off-specular signal, unambiguously related to the lateral heterogeneity of the samples, but also demonstrates clearly that the trilayer lipid structure is preserved. The reflectivity data are also consistent with the presence of a thin ($\sim$ 0.5 nm) and inhomogeneous graphene layer on top of the trilayer (see Fig. 1). It is impossible to extract more quantitative results from these experiments, or to conclude from the effect of the graphene on the lipid layer thickness. But the results are very encouraging and we clearly need to improve our deposition techniques.

3.2 Hybrid graphene-bilayer system

The experimental set-up was switched from solid/air to solid/liquid interface configuration for the last 12 h of beam time where we used to investigate the structure of hybrid lipid-graphene bilayer. This change of configuration was difficult and we lost few hours of beam time to solve some connectivity problems.



(a) (b)

Figure 2: (a) Reflectivity data for an hybrid DSPC-graphe bilayer at 25°C (gel phase) in both H_2O and D_2O. Lines corresponds to the best fit. (b) Corresponding scattering length density profile.

We were able to characterize an hybrid DSPC-graphene-bilayer in gel phase in two contrasts. Unfortunately, we did not have time to characterize the sample in other contrasts (4MW or SMW). It is also important to increase the sample counting statistic in water, which is the most sensitive contrast to the presence of graphene. Nevertheless, the results are very promising: (i) the off-specular signal is very low, indicating that the sample is laterally homogeneous and that the graphene does not affect the bilayer dramatically; (ii) the reflectivity data are compatible with the presence of the graphene bilayer (see Fig. 2).

4 Conclusion

The main objectives of this proposal was to determine with high resolution the structure of hybrid lipid/graphene systems and to understand interaction between different layers. Even though we had difficulties producing large samples, the results we obtained are promising and confirm our previous Raman spectroscopy and ellipsometry experiments. We have obtained important information on supported hybrid graphene-bilayer. This results have to be confirmed and it will be very important to do a full multi-contrast characterization with good statistics to complete our results. We believe that such a characterization, together with our previous Raman spectroscopy and ellipsometry experiments, could lead quickly to a publication, in the framework of Lia Lima Phd to be completed in the fall of 2018. We would like to ask for an EASY ACCESS experiments for this.

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