Experimental report

28/03/2024

Proposal: 8-02-983 **Council:** 10/2022

Title: The role of charge for interactionwith bio-membranes: ¿-Synuclein and Synaptobrevin-2

Research area: Biology

This proposal is a new proposal

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Samples: h-d DMPC/DMPG mebrane + Synaptobrevin(1-96)
h-d DMPC/DMPG mebrane + Synaptobrevin(1-116)
h-d DMPC/DMPG mebrane + alpha synucleine

Instrument	Requested days	Allocated days	From	To
FIGARO	5	0		
D17	5	3	01/04/2023	04/04/2023

Abstract:

In our current research we focus on the interaction of two neuronal IDPs with bio-membranes:¿-Synuclein and Synaptobrevin-2 .
We would like to investigate the interaction of both IDP with membranes of varying charge composition (fraction of negative DMPG in neutral DMPC) to examine the configuration in/at the membrane and in the adjacent solution. For ¿-Syn we want to identify the location of ¿-helices and the configuration of disordered regions.
For Syb2 we will examine the disordered soluble region with and without the transmembrane region .

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In this experiment, the configuration of intrinsically disordered protein alpha-synuclein (α-Syn) in/at differently charged lipid's membrane (fraction of negative DMPG in neutral DMPC) has been investigated.

For the experiment, 3 days of beamtime have been allocated at D17.

Sample	**Protein concentration [mg/ml], total volume [ml]**
DMPC lipid membrane + α-Syn	0.1, 15
25% of DMPG/DMPC lipid membrane + α-Syn	0.1, 15
50% DMPG/DMPC lipid membrane + α-Syn	0.1, 15

Table 1 List of samples measured at D17.

Data have been collected with the following setup parameters:

Angle	DAN	SAN	S2W	S3W	OPENING	TRS
Angle 1	2,8	0,8	1,5	0,75	0,85	0
Angle 2	7,6	3,2	6	3	4	0

Table 2 D17 setup parameters.

An average time of ≈3 hrs was required to measure empty membrane and then ≈5 hrs to measure membrane with the protein for each sample in three different contrasts (H2O, D2O, SMW (silicon-matched (38% D2O/62% H2O) water)). All the data have been collected at T=37°C.

DMPC and 25% DMPG/DMPC lipid bilayers were made by vesicle fusion method.

50% DMPG/DMPC lipid bilayer was made by the Langmuir-Blodgett trough method.

Preliminary results:

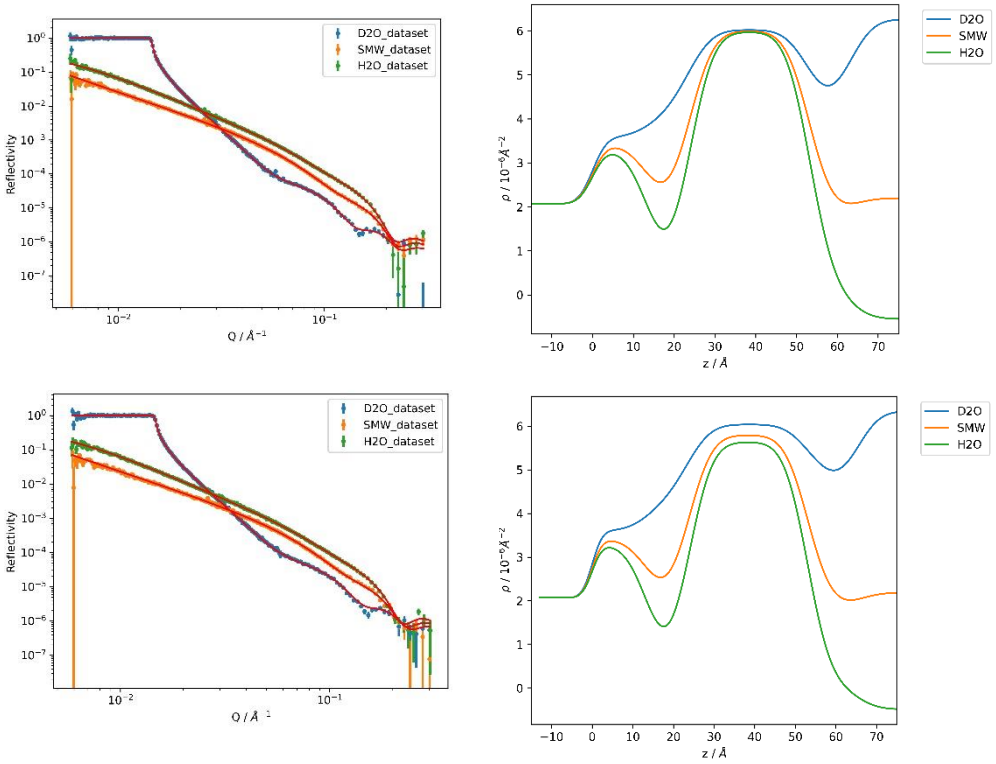

Figure 1 Neutron reflectometry profile and model data fits and the scattering length density profiles these fits describe for deposited DMPC bilayer without (top) and with α-Syn protein (bottom).

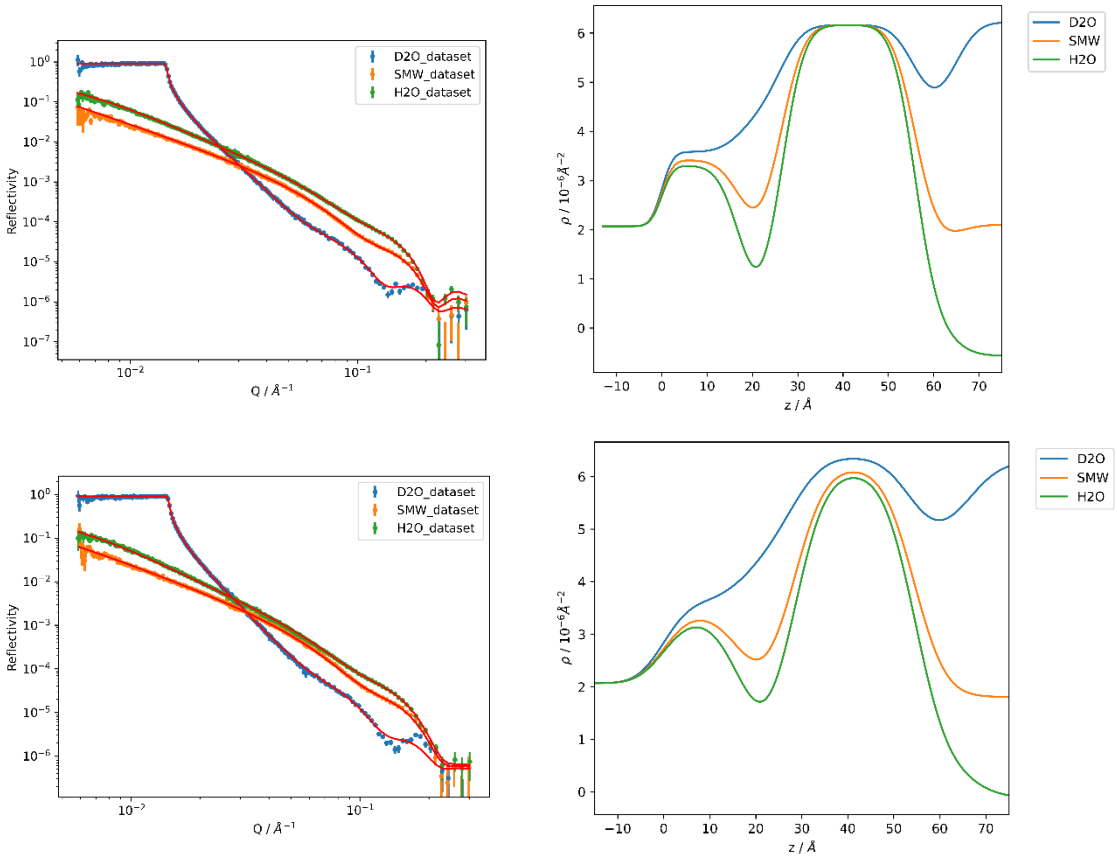

Figure 2 Neutron reflectometry profile and model data fits and the scattering length density profiles these fits describe for deposited 25% DMPG/DMPC bilayer without (top) and with a-Syn protein (bottom).