Experimental report

11/04/2022

Proposal: 9-13-1000 **Council:** 4/2021

Title: Lipoprotein capacity to exchange lipids from normo- and hyper- triglyceridemic individuals or presentinglipoprotein(a)

Research area: Other...

This proposal is a resubmission of 8-05-465

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Samples: Lipids
lipoproteins purified from individuals with distinctive lipid profiles

Instrument	Requested days	Allocated days	From	To
D17	4	3	17/09/2021	20/09/2021
FIGARO	4	0		

Abstract:

The complexity of lipoprotein composition makes it very difficult to map out its role on atherosclerosis development and contradictory results are found in the literature. Not only there are differences in terms of apolipoproteins present in the different lipoprotein classes but also the type of lipids and the proportion between them. Neutron reflection excels at determining the lipoprotein capacity to exchange fats and the exchange results so far mirror clinical data suggesting this model system can be used to explore the function of lipoproteins versus composition. So far we have used pooled samples from healthy adult males but now we are ready to look at differences between individuals with distinctive lipid profiles. In this proposal we aim at finding whether there are differences in lipid exchange between lipoproteins from individuals with normo- and hypertriglyceridemic profiles with or without the presence of lipoprotein(a), a known marker for atherosclerosis. These results will advance our understanding on atherosclerosis development, which is the main killer in the west.

Experimental Report D17 (9-3-1000)

We have studied the interaction between model membranes, Spike protein and HDL. So far we have used HDL pooled samples from healthy adult males but now we wanted to look at differences between individuals with distinctive lipid profiles. In this experiment, our goal was to find out if there are differences in lipid exchange between lipoproteins from individuals with normo- and hypertriglyceridemic profiles with or without the presence of lipoprotein (a), a known marker for atherosclerosis. These results will improve our understanding of the development of atherosclerosis, which is the leading cause of death in the West.

To do this, we prepared supported lipid bilayers composed of deuterated 1,2-dimyristoyl-D54-3-sn-glycerophosphatidylcholine (dDMPC) and perdeuterated cholesterol (dcholesterol) at 80:20 mol% as model membranes and characterized in three isotopic contrasts at 37 ° C using neutron reflection at the D17 reflectometer. After characterization in DTBS, HTBS, and cmSi we incubated six independent model membranes with either: The SARS-CoV-2 Spike protein, HDL (normo- And hypertriglyceridemic), and a mixture of HDL (normo. And hypertriglyceridemic) and SARS-CoV-2 Spike protein, as you can see in the following table.

Table 1. samples used during the incubation.

Surface	CODE	Sample
1	LL	HDL (Low Triglycerides, Low Cholesterol)
2	LH	HDL (Low Triglycerides, High Cholesterol)
3	LH + S	HDL LH + Spike protein
4	HH	HDL (High Triglycerides, High Cholesterol)
5	HH + S	HDL HH + Spike protein
6	LL + S	HDL LL + Spike protein

Figure 1 shows the neutron reflection profiles during the 5 hours of incubation at physiological conditions (HTBS pH 7.4 and 37°C). Through our experiments, we were able to observe that in the presence of the Spike protein, the removal of lipids decreases for the samples composed of HDL LL and HH, as had been previously obtained in previous experiments. However, the HDL LH + Spike sample showed the opposite behaviour. More experiments are needed to understand how the composition of HDL may affect the process of membrane lipid exchange and lipid removal. For future experiments, we hope to study the role of unsaturated lipids in lipid removal.

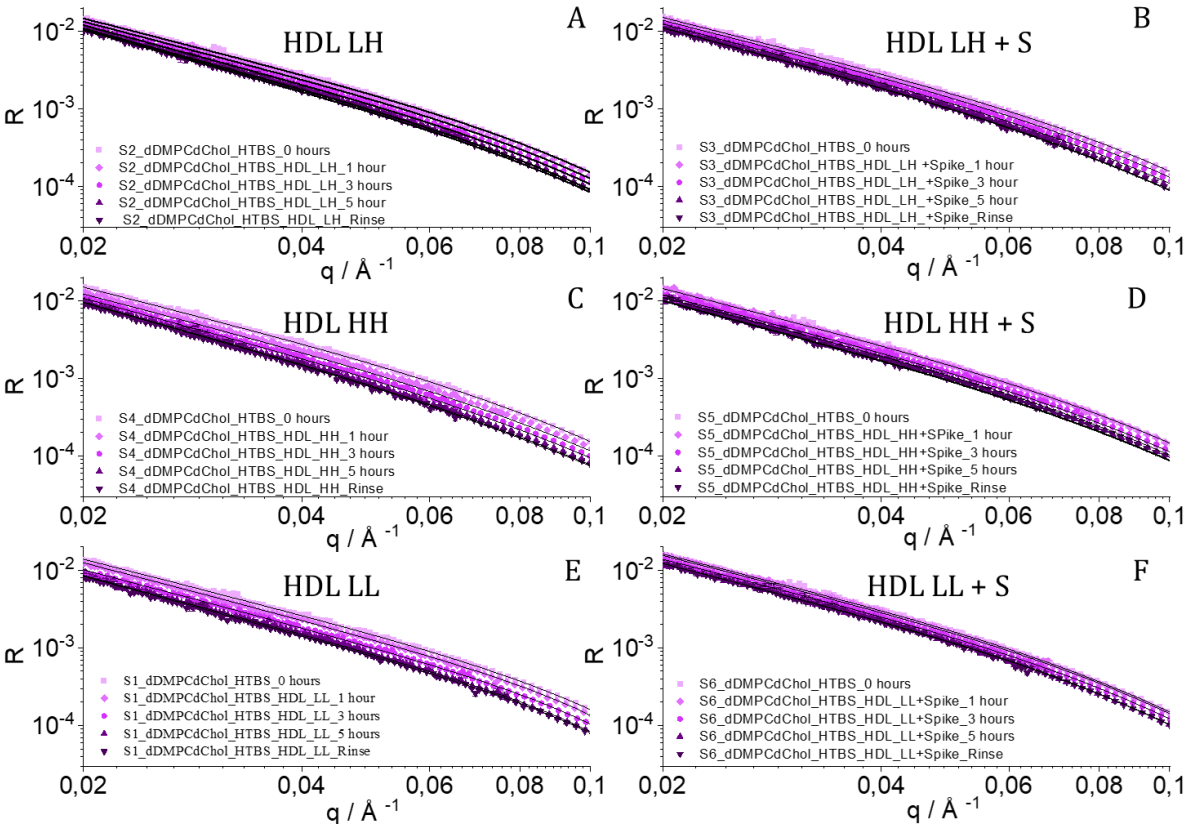

Figure 1. Neutron Reflection profiles of the kinetics including best fits (Line) and raw data (symbol). for the model membranes exposed to HDL (left column, Figures 1, A, C and E) and HDL + Spike protein (right column, Figures 1, B, D and F) in h-TBS at 37 °C after 5 hours of incubation and upon rinsing with excess h-TBS.