Experimental Report

Proposal: **8-02-668** **Council:** 10/2012

Title: Interaction of the beta-amyloid peptide with lipid bilayers: the roleof omega-3 fatty acids.

This proposal is a new proposal

Researh Area: Soft condensed matter

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Samples: Aâ(25–35) amyloid peptide, Gly-Ser-Asn-Lys-Gly-Ala-Ile-Gly-Leu-Met
POPC (C44H84NO8P)
phospholipids, omega-3 fatty acids
Cholesterol (C27H46O)

Instrument	Req. Days	All. Days	From	To
D17	3	3	28/05/2013	31/05/2013

Abstract:

The morphological hallmarks found in the brains of AD patients are extracellular senile plaques, composed of insoluble beta-amyloid peptide (Abeta) fibrillar aggregates. Abeta peptides derive from the proteolytic cleavage of the trans-membrane amyloid precursor protein and the predominant form is Abeta(1-42). Once Abeta is produced, it can be released as a soluble, unfolded unimer into the extracellular environment and be removed or it could accumulate, and eventually self-aggregate in ordered fibrils, undergoing a conformational transition to â-sheet structure. Many studies suggest that the membrane surface may act as a two-dimensional template for fibril nucleation seeds. A particular interest has been developed in Aâ–membrane interactions in order to elucidate the molecular mechanisms of the Aâ-induced cellular dysfunctions underlying the pathogenesis of AD. At the same time, a strong interest is addressed to define strategies for AD prevention and therapy. One of these is related to the dietary components, like omega-3 fatty acids, that appear to play an important role in preventing the disease, possibly changing the physic-chemical properties of the membrane.

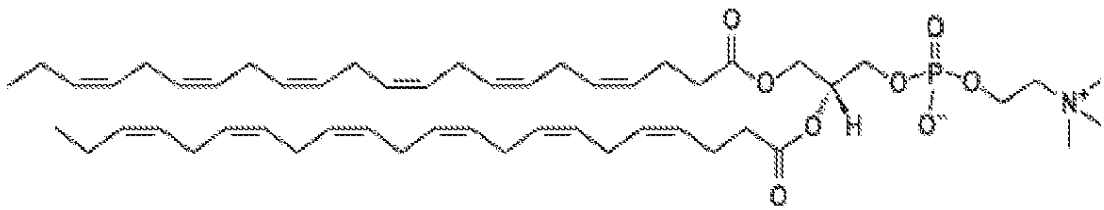
Experimental Report (N. 8-02-668)

Interaction of the beta-amyloid peptide with lipid bilayers: the role of omega-3 fatty acids.

Introduction

Omega-3 fatty acids are polyunsaturated phospholipids which are considered essential for the normal metabolism. The 22:6(cis)PC is the most abundant polyunsaturated acyl chain present in the phospholipid constituting mammalian brain. Some studies suggest that fatty acids may delay or prevent the progression of certain neurodegenerative disorders, like the Alzheimer's disease (AD). Clinical and epidemiological studies have shown that omega-3 fatty acids can affect the risk of the development of AD. AD is characterized by cognitive impairment, intracellular neurofibrillary tangles, synaptic loss, and extracellular β-amyloid plaques. Many evidences have suggested a direct implication of lipid membranes in the mechanisms of fibrillization, favoring the Aβ misfolding and aggregation.[1]

With the present proposal, we have investigated the effect of the omega-3 fatty lipid, 22:6(cis)PC, in palmitoyl oleoyl phosphatidylcholine (POPC) bilayers, mimicking the natural membrane by means of NR measurements.



Molecular structure of 1,2-didocosahexaenoyl-*sn*-glycero-3-phosphocholine (22:6cisPC).

The aim of our research is to investigate the effect of 22:6cisPC in modulating the interaction of the most short active β-amyloid fragment, Aβ(25-35), with biomembranes[2] focusing on their biophysical and micro-structural properties. Basing on a preliminary

study performed by using other different techiniques[3], the idea is to obtain major information from Neutron Reflectivity investigations.

Neutron Reflectivity measurements

NR measurements have been performed on D17 reflectometer, using D_2O, 4MW and H_2O as solvent contrasts. Supported Single Lipid Bilayers (SSLBs) were prepared by vesicles adsorption on silicon bares. Small Unilamellar Vesicles (SUVs), of 25-35 nm in diameter, were formed by vortexing and sonicating for 3×10 min the MLVs suspension. The SUVs suspension (0.5 mg mL^{-1}) was injected into the NR cell, allowed to diffuse and adsorb to the silicon surfaces over a period of 30 min. Afterward the sample cell was rinsed once with deuterated water to remove excess lipid. Peptide solution was added after bilayer formation. We used a synthetic hydrogenated Aβ(25-35) peptide (GSNKGAIIGLM).

We have realized a study on the effect of the omega-3 fatty acid, 22:6(cis)PC, on the microstructure of supported lipid bilayers composed by the hydrogenated-POPC, in the absence and presence of 22:6(cis)PC. The results, shown in Figure 1, indicate that in the absence of omega-3, no peptide-membrane interaction was observed. In contrast, the presence of 22:6(cis)PC favours the peptide-membrane interaction. This result could be related to an effect of the 22:6(cis)PC omega-3 fatty lipid on the micro-structural properties of the biomembranes, influencing consequently the interaction with the Aβ(25-35).

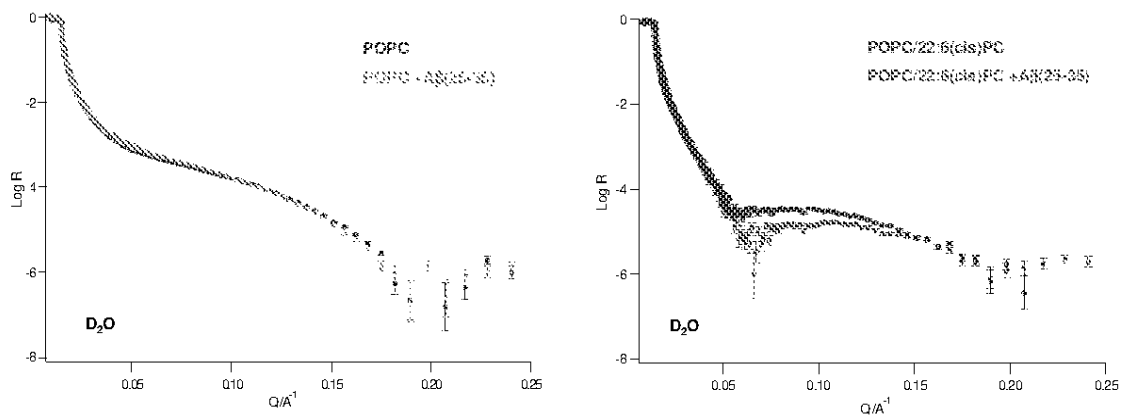


Figure 1 – NR profiles in D_2O for the POPC (on the left) and POPC/22:6(cis)PC in the absence and presence of Aβ(25-35) peptide.

Conclusions

Polyunsaturated omega-3 fatty acids are increasingly proposed as dietary supplements able to reduce the risk of development or progression of the Alzheimer's disease (AD). To date, the molecular mechanism through which these lipids act has not been yet univocally identified. In this preliminary NR investigation, we observed that the omega-3 fatty lipid, 22:6(cis)PC, influences the properties of POPC bilayers, inducing micro-structural changes which can influence the interaction with the Aβ(25-35) peptide.

Other NR experiments could be useful in order to obtain major information on the peptide-bilayers interactions and on the role played by the principal lipids of cell biomembranes.

References

1. Hooijmans C.R., Pasker-de Jongb P.C.M., de Vriesa R.B.M., Ritskes-Hoitinga M. The effects of long-term omega-3 fatty acid supplementation on cognition and Alzheimer's pathology in animal models of Alzheimer's Disease: a systematic review and meta-analysis. *J. Alzheim. Dis.* 28 (2012) 191–209.
2. D'Errico G., Vitiello G., Ortona O., Tedeschi A., Ramunno A., D'Ursi A. M. Interaction between Alzheimer's Aβ(25–35) peptide and phospholipid bilayers: The role of cholesterol. *Biochim. Biophys. Acta* Acta 1778 (2008) 2710–271.
3. Vitiello G., Di Marino S., D'Ursi A. M., D'Errico G. Omega-3 Fatty Acids Regulate the Interaction of the Alzheimer's Aβ(25–35) Peptide with Lipid Membranes. *Langmuir* 29 (2013) 14239–14245.