The Lipidation Profile of Aquaporin-0 Correlates with The Acyl Composition of Phosphoethanolamine Lipids in Lens Membranes

Vian S. Ismail, Jackie A. Mosely, Antal Tapodi, Roy A. Quinlan, John M. Sanderson

PII: S0005-2736(16)30234-6
DOI: doi: 10.1016/j.bbamem.2016.06.026
Reference: BBAMEM 82260

To appear in: *BBA - Biomembranes*

Received date: 26 May 2016
Revised date: 24 June 2016
Accepted date: 30 June 2016

Please cite this article as: Vian S. Ismail, Jackie A. Mosely, Antal Tapodi, Roy A. Quinlan, John M. Sanderson, The Lipidation Profile of Aquaporin-0 Correlates with The Acyl Composition of Phosphoethanolamine Lipids in Lens Membranes, *BBA - Biomembranes* (2016), doi: 10.1016/j.bbamem.2016.06.026

The Lipidation Profile of Aquaporin-0 Correlates with The Acyl Composition of Phosphoethanolamine Lipids in Lens Membranes

Vian S. Ismail,[a,d] Jackie A. Mosely,[a] Antal Tapodi,[c] Roy A. Quinlan[b,c] and John M. Sanderson[a,c]

[a] Chemistry Department, Durham University, Durham, DH1 3LE, UK
[b] School of Biological Sciences, [c] Biophysical Sciences Institute, Durham University, Durham, DH1 3LE, UK
[d] Department of Biology, Faculty of Science, Soran University, Soran, Kurdistan Regional Government, Iraq

Abstract

The lens fiber major intrinsic protein (otherwise known as aquaporin-0 (AQP0), MIP26 and MP26) has been examined by mass spectrometry (MS) in order to determine the speciation of acyl modifications to the side chains of lysine residues and the N-terminal amino group. The speciation of acyl modifications to the side chain of one specific, highly conserved lysine residue (K238) and the N-terminal amino group of human and bovine AQP0 revealed, in decreasing order of abundance, oleoyl, palmitoyl, stearoyl, eicosenoyl, dihomo-γ-linolenoyl, palmitoleoyl and eicosadienoyl modifications. In the case of human AQP0, an arachidonoyl modification was also found at the N-terminus. The relative abundances of these modifications mirror the fatty acid composition of lens phosphatidylethanolamine lipids. This lipid class would be expected to be concentrated in the inner leaflet of the lens fiber membrane to which each of the potential AQP0 lipidation sites is proximal. Our data evidence a broad lipidation profile that is both species and site independent, suggesting a chemical-based ester aminolysis mechanism to explain such modifications.

1. Introduction

Membrane-associated peptides are known to undergo intrinsic lipidation reactions by acyl transfer from membrane lipids [1]. The benchmark peptide for this process is melittin, which is lipidated in synthetic liposomes on the N-terminal amino group and on the side chains of internal lysine and, less commonly, serine residues [2,3]. In the intrinsic lipidation reactions of melittin, little selectivity is found for the aminolysis reaction with the *sn*-1 and *sn*-2 glyceryl esters, and the acyl group distribution of the lipidated products reflects the fatty acyl composition of the liposomal membrane. In principle, membrane-embedded proteins should be susceptible to similar reactions *in situ*, but this is still an open question and the importance of non-ezymatic acylations is still being established [4]. Lipidation events that do not correspond to any of the known consensus sequences for enzyme-mediated modifications and exhibit an acyl group profile that reflects the lipid composition of the proximal membrane leaflet would be the first evidence for non-enzymatic lipidation. The eye lens contains some of the oldest proteins in the mammalian body and integral membrane proteins, such as AQP0, are excellent candidates to test this hypothesis. There are two known lipidation sites in AQP0 one at the N-terminus and the other at Lys-238 (Fig. S1-S3) [5–7]. Neither match consensus sequences for enzymatically mediated lipidation events. *In vitro* palmitoylation of AQP0 has been shown to be a post-translational, rather than a co-translational, event [8]. The longevity of lens proteins such as AQP0 in the plasma membrane of lens fiber cells [7], and the fact that all intracellular organelles are