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Two dimensional VOPBA reveals laminin receptor (LAMRI) interaction with dengue virus serotypes 1, 2 and 3

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Abstract

Background: The search for the dengue virus receptor has generated many candidates often identified only by molecular mass. The wide host range of the viruses *in vitro* combined with multiple approaches to identifying the receptor(s) has led to the notion that many receptors or attachment proteins may be involved and that the different dengue virus serotypes may utilize different receptors on the same cells as well as on different cell types.

Results: In this study we used sequential extraction of PS Clone D cell monolayers with the detergent β -octylglucopyranoside followed by sodium deoxycholate to prepare a cell membrane-rich fraction. We then used 2 dimensional (2D) gel electrophoresis to separate the membrane proteins and applied a modified virus overlay protein binding assay (VOPBA) to show that dengue virus serotypes 1, 2 and 3 all interact with the 37 kDa/67 kDa laminin receptor (LAMRI), a common non-integrin surface protein on many cell types.

Conclusion: At least 3 of the 4 dengue serotypes interact with the 37 kDa/67 kDa laminin receptor, LAMRI, which may be a common player in dengue virus-cell surface interaction.

Background

The dengue viruses have become recognized as important global pathogens causing dengue haemorrhagic fever not only in Southeast Asia but also in South and Central America and in the Caribbean.[1,2]. There are 4 closely related dengue viruses referred to as DENV-1, DENV-2, DENV-3 and DENV-4[3]. They are mosquito borne viruses with a single stranded positive sense RNA genome around 11 kilobases in length, and are able to infect both mosquito and human hosts. A wide range of cell types from multiple species is susceptible to infection with dengue viruses *in vitro*. Numerous studies have attempted to identify the cell surface receptor or receptors utilized by the dengue viruses to gain entry into susceptible cells, but multiple approaches using different cell lines and differ-

ent dengue virus strains have generated many candidate DENV interacting proteins identified in some cases only by molecular mass [4-11]. Heparan sulfates[12] and the C-type lectins DC-SIGN and L-SIGN have been shown to mediate infection by dengue viruses[13] and most recently, studies using a standard virus overlay protein binding assay (VOPBA) have suggested that in the liver cell line HepG2, different DENV serotypes utilize different cell surface molecules[14]. More specifically, mass spectrometric methods have been used to identify reactive bands using VOPBA and it has been suggested that DENV-2 interacts with GRP78[15] while DENV-1 interacts with the 37 kDa/67 kDa high affinity laminin receptor[16].