P13 protein C-terminal peptide and N-terminal peptide of GBV-B forms ion channels in artificial lipid bilayers

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The GB virus B (GBV-B) is a *Flavivirus* that causes acute hepatitis in a number of New World primates. The GBV-B is phylogenetically closely related to the hepatitis C virus (HCV) (Karayiannis & McGarvey, 1995; Erker *et al.*, 1998; Muerhoff *et al.*, 1995). The two viruses share similar genomic structure and organisation (Muerhoff *et al.*, 1995) and have 28% amino acid similarity in their polyproteins (Beames *et al.*, 2001). Because of this similarity it has been suggested that GBV-B may be used as a tool for testing antiviral compounds that may be effective against HCV (Beames *et al.*, 2000), and this possibility has recently been confirmed using HCV NS3 protease inhibitors (Bright *et al.*, 2004). GBV-B encodes a protein p13 (Ghibaudo *et al.*, 2004) that shares sequence similarity to the HCV p7 protein. The HCV p7 protein has been shown to form ion channels that are blocked by amantadine, a long alkyl chain imino sugar derivative, and hexamethylene amiloride (Griffin *et al.*, 2003; Pavlovic *et al.*, 2003; Premkumar *et al.*, 2004). P13 of GBV-B is a polytopic membrane protein of 119 amino acid residues and has four hydrophobic regions that could span a lipid membrane. TM3 and TM4 of p13 share homologies with the two predicted transmembrane regions of p7 and may be functionally equivalent to the region of p7 which forms ion channels. To test the hypothesis that p13 can form an ion channel like p7, peptides corresponding to the TM1/TM2 (p13-N) and TM3/TM4 region of p13 (p13-C) were synthesised and tested for channel activity. We found that both the peptides form cation-selective ion channels in artificial lipid bilayers. The p13-C channels were blocked with 10 μM amantadine (Premkumar *et al.*, 2006). However, the channels formed by p13-N were not blocked by the same concentration of amantadine, suggesting that perhaps the amantadine binding site of the p13 is in the C-terminus of the protein.

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