Anti-herpes simplex virus activities of sulfomannan oligosaccharide PI-88 and disulfated cyclitols

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Abstract

Herpes simplex virus (HSV) initiates invasion of human cells by binding to the cell surface heparan sulfate (HS) glycosaminoglycan chains. This step is mediated by the viral envelope glycoproteins gC and/or gB. Sulfated polysaccharides are compounds that mimic the structure of HS chains, and therefore are capable of inhibiting HSV attachment to and subsequent infection of cells. However the high molecular weight and associated with it poor tissue-penetrating activity have limited potential antiviral application of sulfated polysaccharides in humans. Here we found that the HS mimetic PI-88, a sulfomannan oligosaccharide of low molecular weight, efficiently reduced, in contrast to conventional sulfated polysaccharides, the cell-to-cell spread of HSV. Analogues of PI-88 with chemical modifications based on the introduction of specific hydrophobic/aromatic group(s) at the reducing end of PI-88 oligosaccharide chain showed enhanced capability to inhibit infection of cells and the cell-to-cell transmission of HSV and respiratory syncytial virus (RSV). One of these analogues (denoted 536), prepared by modification of PI-88 with cholestanol group, exhibited in contrast to the parental compound an HSV-inactivating activity. Furthermore, several disulfated cyclitols, identified by screening for an anti-HSV activity of a large number of low molecular weight sulfated compounds, efficiently reduced the cell-to-cell spread of HSV and demonstrated an HSV-inactivating activity.

The second aim of this thesis was to elucidate the molecular basis for viral resistance to PI-88. Variants of HSV type 1 (HSV-1) and type 2 (HSV-2), selected for by virus propagation in cultured cells in the presence of PI-88 were analysed. Many of these variants had a low infectious titer, indicative of a profound impairment in biological activities of the virus in response to continuous pressure from the drug. These variants were substantially resistant to PI-88 presence during their initial infection of cells and/or their cell-to-cell spread. Nucleotide sequence analysis revealed that PI-88 targeted predominantly the viral envelope glycoproteins that comprise mucin-like region(s), i.e., glycoprotein gC of HSV-1 and glycoprotein gG of HSV-2. The deletion of the mucin-like region of HSV-1 gC (amino acids 33-116) or the deletion of whole HSV-2 gG provided the virus with selective advantage to attach to and to infect cells in the presence of PI-88.

In conclusion, we have identified several novel antiviral compounds. One of these compounds, the PI-88 analogue 536, seems to be an attractive candidate for the development of a topical virucide for prevention of genital HSV infections in humans. We have also identified a novel biological function of HSV-2 gG, i.e., its targeting by sulfated oligosaccharides, which suggests involvement of this protein in HSV-2 attachment to cells or in modulation of this step.

Keywords: herpes simplex virus, antiviral drugs, viral glycoproteins, heparan sulfate mimetic, PI-88

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II. **Ekblad M, Adamiak B, Bergefall K, Nenonen H, Roth A, Bergström T, Ferro V, Trybala E.** Molecular basis for resistance of herpes simplex virus type 1 mutants to the sulfated oligosaccharide inhibitor PI-88. Virology, in revision.

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V. **Ekblad M, Andrighetti-Fröhner CR, Bergström T, Banwell MG, Renner J, Kreipl A, Ferro V, Trybala E.** Analogues of sulfated oligosaccharide PI-88 and disulfated cyclitol DSC3 exhibit potent anti-herpes simplex virus and anti-respiratory syncytial virus activities. In manuscript.