Importance of host cell receptor specificity and immune responses in rotavirus pathogenesis

Rotavirus is the main cause of severe infantile gastroenteritis worldwide in humans and a significant animal pathogen. Major determinants of the tropism of rotaviruses for particular hosts and cells include the ability to utilise certain cell surface receptors and evade innate host immune responses. Extraintestinal disease manifestations resulting from rotavirus infection are also increasingly being recognised. For example, this virus plays a role in accelerating progression towards the onset of type 1 diabetes in genetically at-risk children and mice. It is important to evaluate the efficacy and safety of the current childhood vaccines for rotavirus disease occurring both in the gut and extraintestinally.

Rotaviruses are members of the Reoviridae family. The core within the triple-layered, non-enveloped virion contains 11 segments of dsRNA, encoding six structural proteins found in the virion and six non-structural proteins produced inside infected cells that are necessary to complete the replicative cycle. The inner capsid layer consists of virus protein (VP) 6, which encodes rotavirus group and subgroup antigens. Most human and mammalian rotaviruses fall within group A, the subject of this article. The outermost capsid layer comprises spikes

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**Figure 1.** Cellular attachment, entry and replication initiation by rotavirus.
of VP4 embedded in a layer of VP7 (Figure 1). These proteins independently determine serotype specificity, elicit neutralising antibodies and recognise cellular receptors and entry cofactors.

The process of rotavirus attachment and entry into cells is multifactorial, encompassing at a minimum trypsin cleavage of VP4 into subunits VP5 and VP8, followed by VP8 binding to cell surface sialic acids to facilitate major VP4 spike conformational change and VP5 interaction with the cell membrane. Rotavirus tropism relates to the nature of the sialic acid receptor utilised. For example, our studies show that VP8 of animal strains predominantly recognise glycans on gangliosides G\textsubscript{M3} and G\textsubscript{D1a}, whereas human rotavirus VP8 binds G\textsubscript{M1} glycans\textsuperscript{2,3}. This is significant as expression of these gangliosides varies with host and cell type, and they perform different functions within cells. In particular, G\textsubscript{M1} is a marker for lipid membrane microdomains (rafts) that are implicated in rotavirus-cell entry. Thus, G\textsubscript{M1} usage may facilitate human rotavirus cell entry. I discovered that most human and animal rotaviruses additionally utilise the integrin family of integral membrane proteins, notably through VP5 recognition of the \(\alpha_2\beta_1\) integrin and VP7 interactions with \(\alpha_x\beta_2\) integrin\textsuperscript{4}. As for sialic acid recognition, the ability to use integrin receptors correlates with VP4 serotype specificity\textsuperscript{5}, and varies amongst rotaviruses from different hosts. As illustrated in Figure 1, it has been proposed that rotavirus entry occurs into early endosomes, where the low calcium environment promotes VP7 uncoating, VP4 membrane permeabilisation and delivery of the transcriptionally active, double-layered particle into the cytoplasm\textsuperscript{1}.

Initiation of the rotavirus replicative cycle (Figure 1) leads to production of the rotavirus non-structural proteins (NSP). One of these, NSP1, functions as an antagonist of the host innate immune response to RV. NSP1 blocks type I interferon (IFN) induction by inducing the proteasome-mediated degradation of several IFN regulatory factors and the \(\beta\)-transducin repeat containing protein\textsuperscript{6,7}. The latter protein is necessary for activation of the key NF\(\kappa\)B cell signalling pathway. The ability of NSP1 to inhibit IFN responses varies with the virus strain and host species\textsuperscript{8}. Studies of my group (Figure 2) have shown that rotavirus also subverts the signalling cascade of type I IFN, by inhibiting the nuclear accumulation of the signal transducer and activator of transcription molecules, STAT1 and STAT2, which are necessary for production of antiviral proteins\textsuperscript{7}. Although the rotavirus component responsible for STAT inhibition is currently unknown, this property appears to be highly conserved amongst rotaviruses of human and animals. The overall importance of immune response evasion by rotavirus is shown by the cellular growth restriction exhibited by rotavirus strains unable to inhibit IFN induction\textsuperscript{9}.

Rotavirus infection contributes to conditions other than acute gastroenteritis, including autoimmunity in coeliac disease and biliary atresia\textsuperscript{10-12}. Another lifelong autoimmune disease, type 1 diabetes, results from T cell-mediated destruction of insulin-
producing cells within the pancreas. In Australia, 2000 new cases of type 1 diabetes are diagnosed annually, mostly in children. This disease has major long-term health implications and incurs substantial health costs\textsuperscript{13}. Rotavirus was discovered to be a possible viral trigger for progression of children to type 1 diabetes from our studies\textsuperscript{14-15}. We found that Melbourne children at risk of type 1 diabetes were equally as susceptible to rotavirus infection as the overall population, but often responded to infection with increased islet auto-antibody levels, which are markers of progression to diabetes. It was concluded that rotavirus infection may trigger islet autoimmunity and hasten diabetes onset in at-risk children\textsuperscript{15}. Supportive findings have now been reported in Finnish infants, who showed increased islet auto-antibodies after rotavirus infection in association with early introduction to cow’s milk\textsuperscript{17}. One possibility is that molecular mimicry between a rotavirus protein and islet auto-antigens contributes to this process\textsuperscript{18-19}. My group further discovered that infection with monkey or murine rotavirus accelerates type 1 diabetes onset in mice with pre-existing islet autoimmunity\textsuperscript{20}. Murine diabetes acceleration by rotavirus occurs without gastroenteritis or pancreatic replication, so appears to be immune-mediated rather than resulting from direct pancreatic infection\textsuperscript{19}.

Overall, it is clear that the spectrum of rotavirus disease extends beyond acute gastroenteritis, and many factors help determine rotavirus host range and cell tropism, including cellular receptor specificity and the ability to antagonise host immune responses.

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References


Biography

**Barbara Coulson** completed her PhD with Ian H Holmes on rotavirus immunity in neonates, followed by postdoctoral rotavirus research at the Royal Children’s Hospital, Melbourne with Ruth Bishop. In 1991 she was appointed as a Fellow of the NHMRC and has received continuous awards within this scheme since then. She established her laboratory in the Department of Microbiology and Immunology at The University of Melbourne in 1996, where she is an Associate Professor and Reader.