Parechovirus A infection in the intestinal epithelium

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BACKGROUND

PeV-A1 and PeV-A3 (from the *Picornaviridae* family) are the most prevalent PeV-As genotypes. PeV-A1 is mainly causing gastroenteritis in children, while PeV-A3 is linked to severe neurological disease in infants. One of the entry sites for PeV-As is believed to be the gastrointestinal tract. We characterized viral intestinal entry using enteroids derived from fetal material.

METHODS

To allow viral infection 3D enteroids were broken into single cells and seeded into Transwell® inserts where cells differentiate into the different intestinal cell types (see Figure 1). The system was infected with PeV-A1 and PeV-A3, lab-adapted and clinical strains. Replication kinetics were analyzed using quantitative PCR and TCID50. Infected monolayers were imaged using confocal microscopy to confirm infection and determine the cell tropism.

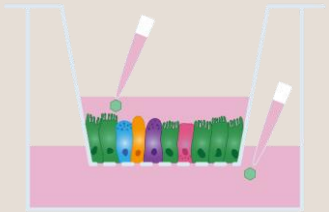

Figure 1. Schematic representation of the intestinal model on a Transwell® insert (showing enterocytes, enteroendocrine, Paneth and goblet cells).

RESULTS

Only lab-adapted strains of PeV-A1 are able to replicate in the model

Both clinical isolates of PeV-A1 and PeV-A3 are able to replicate in the model

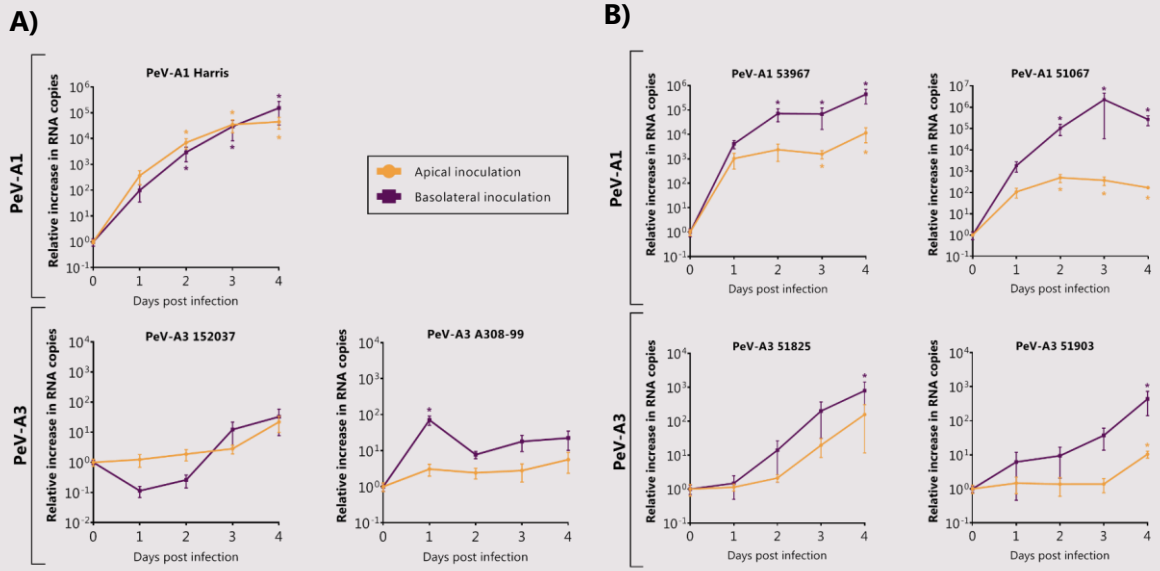

Figure 2. PeV-A1 and PeV-A3 replication kinetics in human intestinal monolayers derived from fetal intestine. Relative increase in RNA copies from apical collection, in both lab-adapted strains (**A**) and clinical isolates (**B**) of PeV-A1 and PeV-A3. Data from three different donors in duplicates.

Infection with clinical isolates of both genotypes generates infectious particles

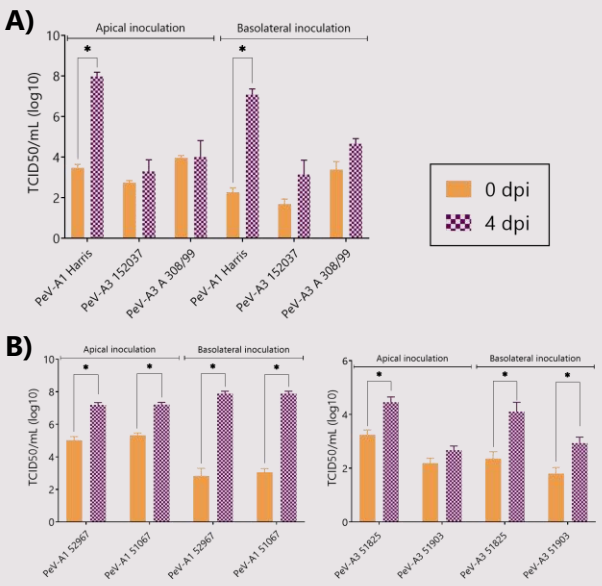

Figure 3. Virus titer at 0 and 4 dpi of apically collected samples for (A) lab-adapted strains and (B) clinical isolates of PeV-A1 and PeV-A3. Data from three different donors in duplicates.

PeV-A1 and PeV-A3 target different cell types

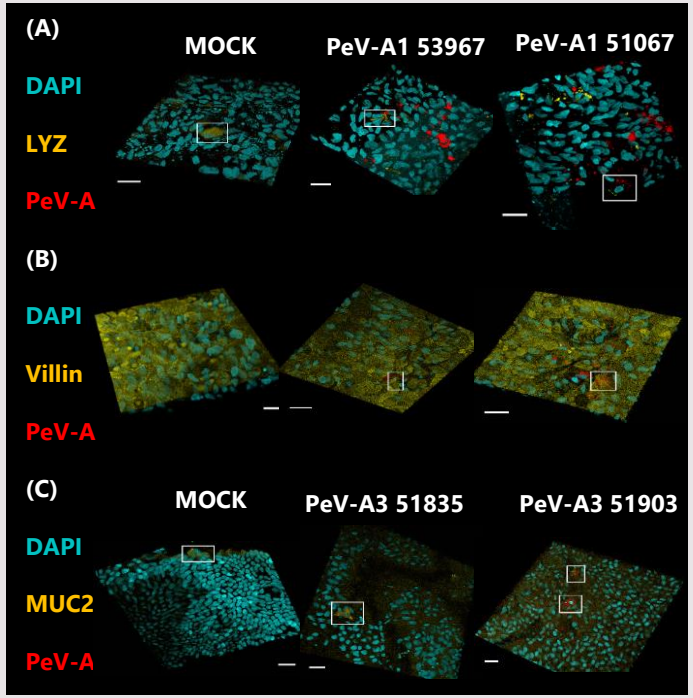

Figure 4. Confocal image z-stacks. Monolayers were stained with enterocyte marker, villin (A), or Paneth cell marker, LYZ (B), or goblet cells marker, MUC2, (C), specific PeV antibody (red) and DAPI for nuclei staining. Scale bars, 20 µm.

CONCLUSION

- PeV-A1 and PeV-A3 can infect the gut organoid model.
- Both genotypes infect preferentially from the basolateral side of the human gut enteroids.
- Different cell tropism is observed, with PeV-A3 infecting goblet cells and PeV-A1 infecting both enterocytes and Paneth cells