

VIDEO LECTURE

EN ESPAÑOL

Let us know what you think FEEDBACK

SEARCH

+ SHARE BOOKMARK PRINT THIS PAGE

<< Previous Next >>

Logo image © Jeffrey Nelson, Rush University, Chicago, Illinois and The MicrobeLibrary

READING:

Murray et al., Microbiology

5th Ed., Chapters 62

## VIROLOGY - CHAPTER SEVENTEEN

### VIRAL AGENTS OF GASTROENTERITIS

# ROTAVIRUSES, CALICIVIRUSES, ADENOVIRUSES, ASTROVIRUSES AND OTHERS

Dr N. Narayan and Dr Helmut Albrecht

A Large number of viruses are found in the human gut; these include some that are associated with gastroenteritis

- Rotaviruses
- Adenoviruses 40/41
- Caliciviruses
- Norwalk-like viruses or small round structured viruses (SRSV)
- Astroviruses
- Small round viruses (SRV)
- Coronaviruses
- Toroviruses

Other viruses found in the gut of a normal individual are not normally associated with gastroenteritis

- Poliovirus
- Coxsackie A virus
- Coxsackie B Virus
- Echoviruses
- Enteroviruses 68-71
- Hepatitis A virus
- Hepatitis E virus
- Adenoviruses 1-39
- Reoviruses

Others are found in the gut as opportunistic infections

- Cytomegalovirus (CMV)
- Herpes simplex virus (HSV)
- Human immunodeficiency virus (HIV)

## ROTAVIRUSES

### Classification

The family *Reoviridae* includes the genus Rotavirus and Coltivirus (includes Colorado Tick Fever virus). Other genera include the Orthreoviruses and Orbiviruses (found in sheep).

Rotavirus was first identified by electron microscopy in 1973 from duodenal biopsies of

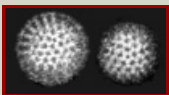


Figure 1. Rotavirus (A double-capsid particle (left), and a single, inner, capsid (right)) © Dr Linda Stannard University of Cape Town, South Africa

children with diarrhea. Human and animal rotaviruses are known.

## Structure

Rotaviruses are non-enveloped viruses with icosahedral symmetry and a double capsid (figure 1). Their electron microscopic appearance shows a 60-80nm wheel with radiating spokes (Latin, rota = wheel) (figure 2).

The rotavirus genome contains double stranded (ds) RNA in 11 segments that can be separated by polyacrylamide gel electrophoresis (PAGE).

### Major structural proteins

Outer structural proteins are VP7 and VP4. VP4 is the viral hemagglutinin and forms spikes from the surface.

Inner core structural proteins are VP 1, 2, 3, and 6. VP6 is an important antigenic determinant.

## Classification

### Groups

There are seven different groups (A to G) based on the antigenicity (each group shares common antigens) and the electrophoretic mobility of their RNA segments. Groups D, E and F have not been found in humans. Group A is the most common and only group A rotaviruses cause human disease in the United States, primarily in the young (under two years of age - infantile gastroenteritis). However, group A rotaviruses can also cause milder diarrhea in older children and adults. Group B has been found to cause human disease in China where there may be annual outbreaks of severe adult and infant diarrhea. More characteristically, group B rotaviruses cause diarrhea in pigs. Group C is found worldwide.

### Serotypes

There are at least 15 different serotypes of rotaviruses. Fourteen G serotypes are based on G protein (GP7) differences. Five predominant strains in the United States (G1, G2, G3, G4, G9) account for 90% of isolates and strain G1 accounts for 73% of infections.

There are 20 P serotypes based on the P protein (VP4) with P4 and P8 predominating.

Common P/G combinations are P8G1, P8G2, P4G2 and P8G4

## Properties

Rotaviruses are stable in the environment for many months and are relative resistant to hand washing. They are susceptible to agents such as 95% ethanol, formalin and "Lysol". They are also unstable to pH below 2.

## Pathogenesis

Affected host cells are mature enterocytes lining the middle and upper end of the intestinal villi. In laboratory animals, hepatocytes are also infected. The infectious particle is thought to be an "intermediate sub-viral particle" (ISVP).

The viral attachment protein is probably exposed after protease digestion in the GI tract removes some or all of the outer capsid protein (VP4). Rotaviruses replicate in the host cell cytoplasm. Virions enter the host cell by endocytosis and viral mRNA is transcribed using the viral RNA polymerase that is already present in the virion to form structural protein units of the capsid. The mRNA segments are assembled into the immature capsid and then replicated to form the double stranded RNA genome. Large amounts of viral particles are shed in diarrheal stools.

Histopathology of infected intestines shows villous atrophy and blunting, due to death of the mature enterocytes and infiltration of lamina propria with mononuclear cells. Subsequently there is repopulation of the villous tips with immature secretory cells (crypt

## WEB RESOURCES

[Rotavirus structure](#)  
(Dr Linda Stannard)

[Gastroenteritis Fact Sheet](#)  
CDC

[Rotavirus Diarrhea in the  
Child Care Setting](#)  
CDC

[Rotavirus](#)  
CDC pdf file

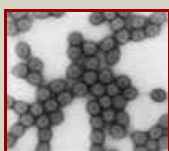
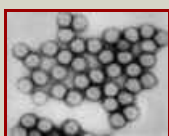


Figure 2.

Transmission electron  
micrograph of intact rotavirus

particles, double-shelled. Distinctive rim of radiating capsomeres. CDC/Dr. Erskine Palmer

hyperplasia).

Cell dysfunction and death results in a net secretion of intestinal fluid, hence the watery diarrhea. Activation of the enteric nervous system may also play a role.

Repopulation with immature secretory cells may contribute to the secondary lactose intolerance that is sometimes seen.



Figure 3A  
Estimated global distribution of the annual deaths caused by rotavirus diarrhea. CDC

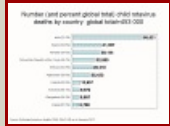


Figure 3B  
National estimates of rotavirus attributable deaths among children under five years of age (2008) WHO

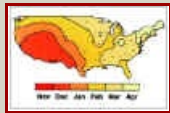


Figure 4.  
Average time of peak rotavirus activity in the contiguous 48 states, United States, July 1991 to June 1997 CDC

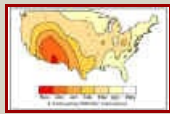


Figure 5.  
Month of peak rotavirus activity — United States, July 1996-June 1997

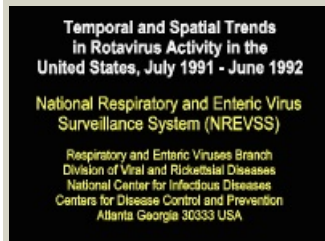
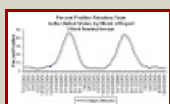


Figure 6. Average time of peak rotavirus activity in the contiguous 48 states, United States, July 1991 to June 1992. This contour plot was derived using the median value for time of peak activity for each laboratory. CDC

Higher resolution movie of above image [avi file](#)

Peak month for reports of rotavirus infections across the US, 1991-97 [avi file](#)



Weekly reports of rotavirus in the US. Seasonal variation. CDC

## Epidemiology

### Distribution

Rotaviruses are found worldwide, causing major gastroenteritis and diarrhea-associated hospitalization and over half a million deaths per year in children under five years of age. According to WHO, five countries (India, Nigeria, the Democratic Republic of the Congo, Ethiopia and Pakistan) accounted for more than half of all rotavirus disease deaths under age five in 2008.

Symptoms include: fever, vomiting, diarrhea and abdominal pain. Seroprevalence studies show that antibody is present in most infants by age 3 years.

Prior to the introduction in the United States of widespread vaccination in 2006, there were up to three million cases of rotavirus infection per year. In about 1 to 2.5% of cases, there was severe dehydration. This resulted in 20 to 60 deaths of children under five each year. In addition, there were 50,000 to 70,000 hospitalizations and over 500,000 visits to doctors' offices per year.

Since the introduction of vaccination there has been a drop in rotavirus-related hospitalizations by up to 86 percent. It is likely that vaccination has also protected non-vaccinated infants by limiting circulating infection. Deaths have also been markedly reduced. In 2008, there were an estimated 14 deaths from rotavirus disease in the United States and fewer than 10 in the United Kingdom compared to 98,621 in India.

### Seasonality

In the U.S.A., rotavirus infections occur in the winter months (November through May). The disease spreads across North America from the warmer climates, starting from Mexico and SW USA and gradually progressing N/NE to reach East Coast and Canada in spring (figures 4 - 6). As might be expected, rotavirus infections are seen year round in the tropics.

### Incubation period

This is thought to be less than 4 days

### Contagious Period

The patient is contagious from before the onset of diarrhea to a few days after the end of diarrhea.

### Age of infections

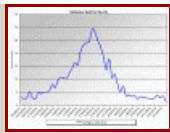
Rotaviruses infect children at a young age. Older infants and young children (4 months - 2 years) tend to be more symptomatic with diarrhea. Young infants may be protected due to trans-placental transfer of antibody. Asymptomatic infections are common, especially in adults. Many cases and outbreaks are **nosocomial**

Group A infections are most common.

Group B has been associated with outbreaks in adults in China

Group C is responsible for sporadic cases of diarrhea in infants around the world.

Spread is mainly person to person via fecal - oral route and through **fomites**. Spread by food and water is also possible. There has been speculation that rotaviruses may also spread via the respiratory route.



Rotavirus  
infections weekly trends  
2012-2013  
CDC

High numbers of viral particles are shed in diarrheal stools ( $10^{10}$ /gm). Infective dose is only 10-100 pfu.

### Clinical Features

Fever can be high grade ( $>102^{\circ}$  F in 30% of patients) and vomiting and nausea precedes diarrhea. Diarrhea is usually watery (no blood or leukocytes), lasting 3-9 days, but longer in malnourished and immune deficient individuals. Necrotizing **enterocolitis** and hemorrhagic gastroenteritis is seen in neonates. *Dehydration is the main contributor to mortality.* Secondary malabsorption of lactose and fat, and chronic diarrhea are possible.

### Diagnosis

Rapid diagnosis can be obtained by antigen detection in stool using ELISA (which uses a monoclonal antibody) and LA. Several kits are commercially available. These detect only Group A rotavirus. Electron microscopy also detects non-Group A viruses.

Group A rotaviruses can be cultured in monkey kidney cells.

Epidemiologic studies use patterns of viral RNA migration by gel electrophoresis (electropherotyping). Different genetic strains may circulate in a given community.

### Treatment

Treatment is just supportive care with rehydration (oral / intravenous). Antiviral agents not known to be effective

### Prevention of spread

Good hand washing technique is important. In addition, surfaces, toilets and toys should be disinfected. Adequate chlorination of water can prevent spread in the community.

### Immunity

Antibodies against VP7 and VP4 are partially protective but the initial infection does not lead to permanent immunity and reinfection can occur at any age. However, subsequent infections are usually less severe than the primary infection.

### Vaccine

Reassortant vaccines are created by genetic reassortment in which non-human rotavirus strains express the antigens of human rotaviruses on their surface. The non-human strains replicate but do not cause disease and are of low pathogenicity in humans.

A live, tetravalent rhesus-human reassortant vaccine (*Rotashield* - Wyeth Laboratories) was first licensed for use in infants in August 1998. It contained human G types 1, 2, 4, and simian G type 3. However, post-licensure surveillance indicated a possible relationship between the occurrence of **intussusception** 3 to 20 days after the vaccine was administered, especially the first dose (15 cases/1.5 million doses were reported). Use of the vaccine was suspended and it was eventually removed from the market in October 1999, when studies confirmed the link between vaccination and intussusception.

*RotaTeq* (Merck) is a live oral vaccine licensed in the United States in 2006. It contains five reassortants (WC3 bovine rotavirus strain with surface proteins of the G1-4 and P1A human serotypes). It does not contain preservatives or thimerosal. Three doses are given at 2, 4 and 6 months of age with the minimum age for the first dose of 6 weeks. The series should not be initiated after 12 weeks. The efficacy of the RotaTeq vaccine is high with 98% reduction in *severe* rotavirus gastroenteritis within the first year of vaccination and a 96% reduction in hospitalization rate. There is also a 74 and 71% reduction of rotavirus gastroenteritis within the first and second years after vaccination.

RotaTeq and  
Kawasaki  
syndrome

*Rotarix* (Avant Immunotherapeutics/Glaxo) is a live, attenuated, monovalent vaccine that contains the G1P[8] human rotavirus strain. It was licensed in the United States in 2008. It has been studied in South America and has a two dose schedule of administration. There is no increase in intussusception. After two doses, there is protection through the first two years of life. Hospitalizations are reduced by 96% and severe rotavirus gastroenteritis by 90%. The vaccine is also effective against rotavirus gastroenteritis of any severity (79%). Significant protection was demonstrated against severe rotavirus gastroenteritis during two rotavirus seasons caused by types G1 (96%), G2 (86%), G3 (94%), G4 (95%), and G9 (85%). These are the most commonly circulating rotavirus types in the United States.

## SMALL ROUND RNA VIRAL AGENTS CAUSING GASTROENTERITIS

This group of RNA viruses morphologically is subdivided into 2 sub-groups:

- Structured - Small round structured viruses (SRSV), Calicivirus, Astrovirus
- Other small viruses that are relatively structureless or featureless - W (Wollan) and Ditchling.

## CALICIVIRUSES

Human caliciviruses were first described in 1976. They belong to the family *caliciviridae* and are non-enveloped, single strand, positive sense RNA viruses. They are 27 to 35 nm in size (figure 9). They appear round in shape with icosahedral symmetry and contain a single capsid protein. The viral surface has 32 cup-shaped depressions ('calici' = chalice or calyx i.e. cup-like) described as the 'Star of David' appearance. Otherwise they are similar to Norwalk group of agents.

## CLASSIFICATION

Caliciviruses can be divided into:

- Norwalk and "Norwalk-like" viruses (NLV)
- "Sapporo-like" viruses (SLV)
- Vesiviruses
- Lagoviruses

NLV (Noroviruses) include:

- Norwalk virus
- Hawaii virus
- Snow Mountain virus
- Montgomery County virus
- Taunton (England) virus

SLV (Sapoviruses) include:

- Sapporo virus
- Manchester virus
- Houston/86
- London/92

New types are named after the place where they were first isolated in relation to outbreaks of diarrhea.

## NORWALK VIRUS AND NORWALK-LIKE VIRAL AGENTS

Norwalk virus was first detected in stools of patients with gastroenteritis (winter vomiting disease) in Norwalk, Ohio in 1968. They cause 40 per cent of non-bacterial gastroenteritis epidemics. Forty five per cent are food-borne and 52 per cent are raw shell-fish associated. They tend to cause rapid (explosive) epidemics in places of close contact such as cruise ships, nursing homes, hospitals and camps. In the electron microscope, these viruses are 27 - 32nm in size with a ragged surface.

## Epidemiology



Figure 7.

Norwalk virus from stool sample from an individual with gastroenteritis.

F.P. Williams, U.S. Environmental Protection Agency



Figure 8.

Typical morphology of Norwalk-like viruses seen by transmission electron microscopy.

The individual virions have a diameter of only 27nm. Wadsworth Center of the New York State Department of Health.

Figure 9. Bovine

calicivirus © 1994 Veterinary Sciences Division - Dr Stewart McNulty at Veterinary Sciences, Queen's University, Belfast.

## WEB RESOURCES

Norwalk Virus Fact Sheet  
CDC

Noroviruses are found world-wide and cause more than 23 million cases of gastroenteritis very year in the United States. They are the cause of more than half of gastroenteritis cases in the US. From seroprevalence studies, it has been found that most people have been infected by the age of four.

There are asymptomatic infections in which the patient is infectious, has seroconverted and sheds virus. The infective dose may be very low (~10pfu) and virus may continue to be secreted during the convalescent period. Protective immunity is short-lived.

### Clinical Features

Adults and children are affected. The infection has a relatively short incubation period of about 24 hours with a range of 12 to 96 hours. The resulting illness is short (less than 3 days). The most prominent symptoms are vomiting, nausea, abdominal cramping and watery diarrhea accompanied by headache, fever and malaise. The 1 to 3 day period of diarrhea is less than that associated with rotavirus infections.

### Treatment

The symptoms are treated by rehydration and the use of anti-diarrheals. Complications are rare but can be found in the immunocompromized.

### Spread

Norwalk virus is spread via the feco-oral route and, perhaps, also through vomit. Outbreaks spread through fecally-contaminated food or water. Norwalk viruses can survive for several days on plastic surfaces such as counter tops and telephones and in water that is chlorinated at the usual levels (up to 10 ppm). They can survive freezing and heating to 60 degrees C. They also survive in steamed shellfish.

### Diagnosis

Stool specimens, vomit, suspected food and environmental swabs (during an outbreak) may be tested using PCR (in state laboratories). Immune electron microscopy is less used. Serology may be used for epidemiologic purposes.

### Control

CDC recommends disinfection of surfaces using bleach (1 part bleach to 50 parts water). Hand sterilization is also important during an outbreak.

## ASTROVIRUSES

Astroviruses were described in relation to an outbreak of gastroenteritis in 1975. They are small single stranded RNA, non-enveloped virus, about 27 - 32nm in size. They are round with an unbroken surface (unlike indented surface of calicivirus) (figure 10 - 12). Their appearance in the electron microscope is a 5 or 6 pointed star within a smooth edge. They contain 3 structural proteins and their genome has been sequenced.

Astroviruses are immunologically distinct from Norwalk and other Caliciviruses - they belong to the family *Astroviridae*

Eight human serotypes are known and there are also animal strains.

### Clinical Features

Infants, children, immunocompromized patients and the elderly are most often affected by astrovirus infections. The incubation period is short (1 to 4 days) and is followed by watery diarrhea, abdominal cramps, headache, nausea, low-grade fever, vomiting (the latter being less common).

### Epidemiology

Astroviruses are endemic worldwide, mainly in children less than 7 years of age. Presently, the true disease burden is unclear. Transmission is person-to-person via fecal-oral route and outbreaks due to fecal contamination of sea-food or water often occur.

### Diagnosis

Electron microscopy and immuno-electron microscopy are especially useful since the virus is often shed in large amounts in stool. Immunofluorescence microscopy detects all



Figure 10.

Astrovirus © 1994 Veterinary Sciences Division - Dr Stewart McNulty at Veterinary Sciences, Queen's University, Belfast.

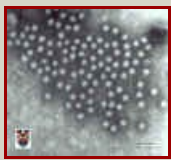


Figure 11.

Astrovirus © 1994 Veterinary Sciences Division - Dr Stewart McNulty at Veterinary Sciences, Queen's University, Belfast.

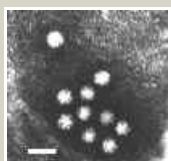


Figure 12.

Human astrovirus US Environmental Protection Agency

## WEB RESOURCES

Viral Agents of Gastroenteritis Public Health Importance and Outbreak

serotypes. ELISA and PCR are also used.

## ADENOVIRUSES

**Adenoviruses** were first isolated in 1953 from adenoidal tissue. The double stranded DNA viruses about 70 to 75nm in diameter. Mammalian adenoviruses belong to the genus *mastadenovirus*. There are six sub-genera of human adenoviruses (A to F) with 51 serotypes some of which have known oncogenic potential. In the laboratory, adenoviruses have found use in gene therapy and vaccine delivery.

Adenovirus serotypes implicated in gastroenteritis are 40 and 41 which belong to serogroup F. They cause diarrheal disease in infants and children less than 4 years of age. These ubiquitous viruses are found in the population year-round and are spread by the feco-oral route. They are not shed in the nasopharynx.

The incubation period of adenoviral gastroenteritis is 3 to 10 days and diarrhea lasts 10 to 14 days; prolonged diarrhea often seen with type 40 infections. This can also lead to intussuseption, mesenteric adenitis and appendicitis.

Isolation requires a special medium, Graham 29.

Diagnosis is made by latex agglutination and ELISA tests or by electron microscopy.

## TOROVIRUSES

Toroviruses (figure 13) belong to the family *Coronaviridae* and the genus *Torovirus* They are pathogens for both humans and animals. They are pleiomorphic, coated, single positive strand RNA viruses. In the electron microscope they have a doughnut shape (torus). They cause watery diarrhea in infants of 2 to 12 months. They are usually diagnosed by electron microscopy.

## CYTOMEGALOVIRUS

These are **herpes viruses** which in normal people give rise to a number of diseases, particularly infectious mononucleosis in western countries. In the immunocompromized, they lead to retinitis, hepatitis and colitis.



[Return to the Virology section of Microbiology and Immunology On-line.](#)



[Return to the Home Page of Microbiology and Immunology On-line](#)

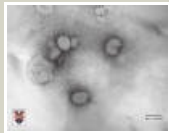


Figure 13  
Torovirus negative stain  
electron microscopy. ©  
Stewart McNulty, Queens  
University, Belfast.