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Chapter 45

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Diarrhoea Caused by Viruses

The gastrointestinal tract is the commonest portal of entry for a variety of pathogens, including viruses, but not all these viruses are causally associated with diarrhoeal disease. Among the viruses that infect enterocytes, or at least use them as a portal of entry, there are two major groups. The first group comprises those viruses that cause systemic infections after entering into the body through the gastrointestinal tract, and diarrhoea, if ever present, is not a major feature of infection. This group includes many enteroviruses, including poliovirus and coxsackieviruses, hepatitis A and E viruses, and some adenoviruses. The second group comprises the viruses that infect the upper small intestine and cause non-inflammatory diarrhoea. It is generally perceived that the enteropathogenic viruses do not normally cause systemic infection. While these viruses are difficult to grow in cell culture, there are often enormous numbers of virions shed into stool, which can be identified by direct electron microscopy or immune electron microscopy. There are currently five genera of viruses recognized as established causes of gastroenteritis in humans, i.e. *Rotavirus*, *Norovirus*, *Sapovirus*, *Astrovirus*, and group F adenovirus.

ROTAVIRUS

Human rotavirus was first discovered in 1973 on thin-section electron microscopy of duodenal biopsies from a child with acute gastroenteritis, and named duovirus.[1] The virus was subsequently found in large numbers in faeces as demonstrated by direct negative-stain electron microscopy[2] and significant antibody titre rises were shown between acute and convalescent sera from diarrhoeal children by immune electron microscopy.[3] The virus was named rotavirus because of its characteristic wheel-shaped (*rota* is Latin for a wheel) morphology on electron microscopy (Figure 45.1).

Geographical distribution

Virtually all children are infected with rotavirus (group A rotavirus) by the age of 3–5 years, whether they live in developing or developed countries.[4,5] Thus, rotavirus is distributed evenly across the world. However, the consequences of infection are markedly different depending on where the child lives, and the majority of deaths due to rotavirus diarrhoea occur in the developing countries of the Indian subcontinent and sub-Saharan Africa (Figure 45.2).[5,6]

Epidemiology

Rotavirus diarrhoea occurs at an earlier age in children in developing countries than in children in developed countries (Figure 45.3).[7] The median age of children hospitalized with rotavirus diarrhoea in many African and Asian counties is 6–9 months, and up to 80% are less than 1 year old.[8] In contrast, the median age in developed countries is 13–16 months and the highest proportion of cases occurs in the second year of life.[7] Nevertheless, in both developing and developed countries, rotavirus is the major cause of severe gastroenteritis requiring hospitalization and, where access to medical intervention is limited, of death. It has been estimated that, from 1986 to 1999, a median of 22% (range 17–28%) of acute diarrhoea cases in children less than 5 years of age were due to rotavirus,[5] but this proportion has nearly doubled recently (from 2000 to 2004) to become 39% (range 29–45%).[6] The estimated annual global mortality due to rotavirus diarrhoea among children less than 5 years of age has also been increased to 611 000 (range 454 000–705 000), reflecting the increasing detection rate of rotavirus as the cause of severe diarrhoea.[6] Included among countries where the disease burden is estimated to be the highest are Afghanistan (1 : 90 children), Democratic Republic of Congo (1 : 130) and Nigeria (1 : 140), and the cumulative incidence of rotavirus diarrhoeal deaths in developing countries is estimated to be on average 1 : 250.[9] On the other hand, in developed countries, hospitalization due to rotavirus in children under 5 years of age is estimated as between 1 : 20 and 1 : 80.[7] In temperate countries, rotavirus infections peak in the winter and early spring, with fewer cases at other times. In tropical countries, rotavirus infections occur throughout the year, although more cases are observed in the cooler and drier months (Figure 45.4).

Virology

Rotavirus is a genus within the family Reoviridae, and within the genus there are seven groups (A to G), each of which represents a separate species, e.g. *Rotavirus A*, *Rotavirus B*, etc. Only group A, B and C rotaviruses are established as human pathogens. Group A rotavirus has much greater medical importance and, unless men-

tioned otherwise, rotavirus usually means group A rotavirus. Group B rotavirus infection is rare and affects both adults and children, causing both outbreaks and sporadic infections, primarily in China, India and Bangladesh.[10,11] Group C rotaviruses tend to affect older children than group A rotavirus, and up to a third of adult humans have serological evidence of infection with group C rotavirus.[12,13]

By conventional negative-stain electron microscopy, rotavirus has a characteristic double capsid structure measuring approximately 75 nm in diameter (Figure 45.1), but cryoelectron microscopic studies have shown that a rotavirus virion consists of a triple-layered capsid with 60 spikes protruding from its surface, making its overall diameter nearly 100 nm. As shown in Figure 45.5, the outermost layer (outer capsid) consists of proteins, VP4 and VP7, each of which independently serves as a neutralization antigen. The serotype defined by the VP4 protein is called the P type, for protease-sensitive protein (because VP4 is proteolytically cleaved into VP8* and VP5*), and the serotype defined by the VP7 protein is called the G type, for glycoprotein. The inner capsid or the middle layer consists of the most abundant viral protein, VP6, which is the major protein against which antibodies are raised during infection with rotavirus. These antibodies are, however, non-neutralizing. The core or the innermost layer consists of VP2, a scaffolding protein, and inside this layer are VP1 (viral RNA-dependent RNA polymerase) and VP3 (guanyltransferase), which is present in association with the 11 segments of double-stranded genomic RNA. In addition to these five structural proteins, there are six non-structural proteins (NSPs), each of which is encoded by a single genome segment, except for NSP5 and NSP6, which carry out various functions during replication and morphogenesis. NSP4 works as a chaperone protein enabling the subviral particle to acquire the outer capsid proteins VP4 and VP7 during the later phases of viral morphogenesis. NSP4 also acts as viral enterotoxin, causing diarrhoea in newborn mice.[14–16]

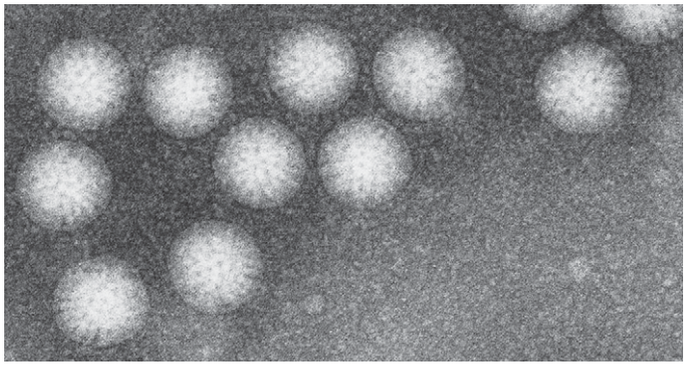

Figure 45.1 Negative-stain electron micrograph of rotavirus particles. (×200 000.)

Rotavirus genomic RNA can be extracted directly from clinical specimens and separated by polyacrylamide gel electrophoresis (PAGE). With this, two major RNA migration patterns are recognized in which genome segments 10 and 11 of long RNA pattern

Figure 45.2 Map showing global distribution of rotavirus mortality in children less than 5 years of age. Each dot represents 1000 deaths. (Reprinted from Parashar et al.[6])

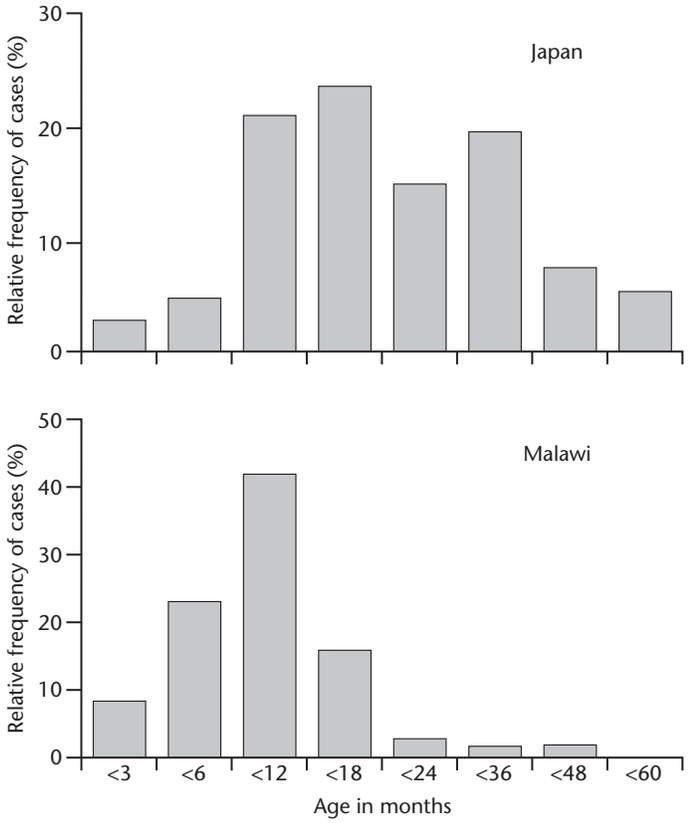

Figure 45.3 Two contrasting patterns of age distribution of rotavirus diarrhoea occurring in Malawi (as an example of a developing country) and in Japan (as an example of a developed country). There is a clear difference in the median ages of children hospitalized with rotavirus diarrhoea in the two countries. (Data taken from Nakagomi et al.[7] and Cunliffe et al. [unpublished].)

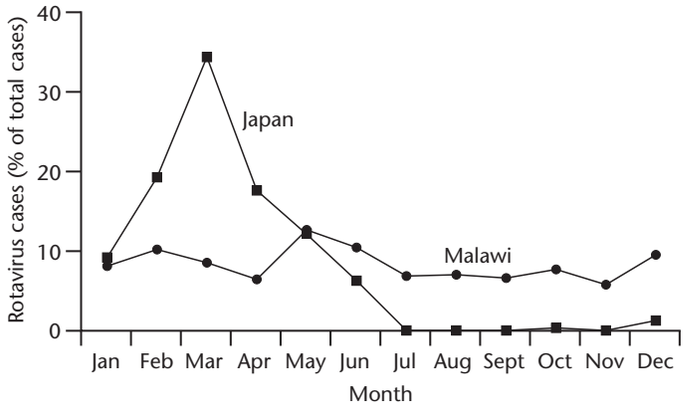

Figure 45.4 The contrasting seasonality of rotavirus diarrhoea occurring in Malawi (as an example of a country in the tropics) and in Japan (temperate climate). Rotavirus gastroenteritis peaks in winter to early spring in temperate countries, while the disease is year-round in the tropics. (Data taken from Nakagomi et al.[7] and Cunliffe et al. [unpublished].)

viruses migrate faster than do those of short RNA pattern viruses (Figure 45.6).[17] The precise migration pattern is characteristic for each rotavirus strain and is called an 'electropherotype', which has been extensively used in molecular epidemiological studies.[18]

The serotype is the most important antigenic determinant of rotavirus and is defined traditionally by serological assays. However, serological assays are now being replaced by molecular typing. While there is an exact correlation between G serotype and G genotype, thereby allowing the use of the same numbering system, different numbering systems are adopted to designate P serotype and P genotype. In the latter, a number for P genotype is designated within a squared bracket. Thus, the serotype of prototype human rotavirus strain Wa is described as G1P1A[8]. There are currently 16 G serotypes and 26 P serotypes described in the literature, but the G and P type combinations (Figure 45.7) detected in human rotaviruses are mostly limited to G1P[8], G2P[4], G3P[8], G4P[8] and G9P[8].[19,20] However, previously rare G[12] strains appear to have emerged across the world,[17,21] and G8 strains with either P[6] or P[4] account for a significant proportion of human rotavirus strains in Africa.[22] Such genetic diversity seems to be generated by frequent reassortment of the genome segments and interspecies transmission of rotaviruses between humans and animals.[23,24]

Further discrimination between group A rotaviruses is based on subgroup (I, II, I+II, neither I nor II) antigens that are carried on the VP6 protein, and subgrouping rotavirus isolates is sometimes used in epidemiological studies.

Pathogenesis

Large amounts of rotavirus (up to 10^{11} virus particles per gram) are excreted in faeces during the acute phase of infection, and the shedding continues after the symptoms cease, sometimes for more than 1 month, albeit detectable only by sensitive reverse-transcription (RT) PCR assays.[25] Children with severe diarrhoea excrete more virus than do children with less severe diarrhoea.[26] The minimum infective dose is as low as 10^2–10^3 virus particles in adult volunteers.[27] Person-to-person spread by the faecal–oral route is most likely, but there are possibilities for air-borne and water-borne transmission of rotavirus. The incubation period is usually 1–3 days. Rotavirus exclusively infects the mature differentiated villous enterocytes of the small intestine. Unlike parvovirus, rotavirus cannot infect the immature villous crypt cells, hence stem cells are spared, nor does rotavirus infect colonic enterocytes. Rotavirus attaches to its cellular receptors (sialoglycoprotein and integrins) via the VP4 protein, but whether the virus enters the cells by endocytosis or direct penetration has not been determined. Three mechanisms have been described for the pathogenesis of rotavirus diarrhoea. In the first 12–24 hours post infection, enterocytes are intact but levels of the brush border disaccharidases (sucrase, maltase, lactase) are greatly decreased.[28] This is apparently due to interference with transport of the enzymes to the brush border.[29] As a result, disaccharides in the diet cannot be hydrolysed to monosaccharides and thus cannot be absorbed, producing an osmotic diarrhoea. Second, NSP4 has an effect in opening calcium channels in the enterocyte. This causes an efflux of sodium and water, and a secretory diarrhoea.[14] Finally, the raised intra-enterocyte calcium concentration causes enterocytes to die by oncosis.[30] The rate of death of the mature villous tip

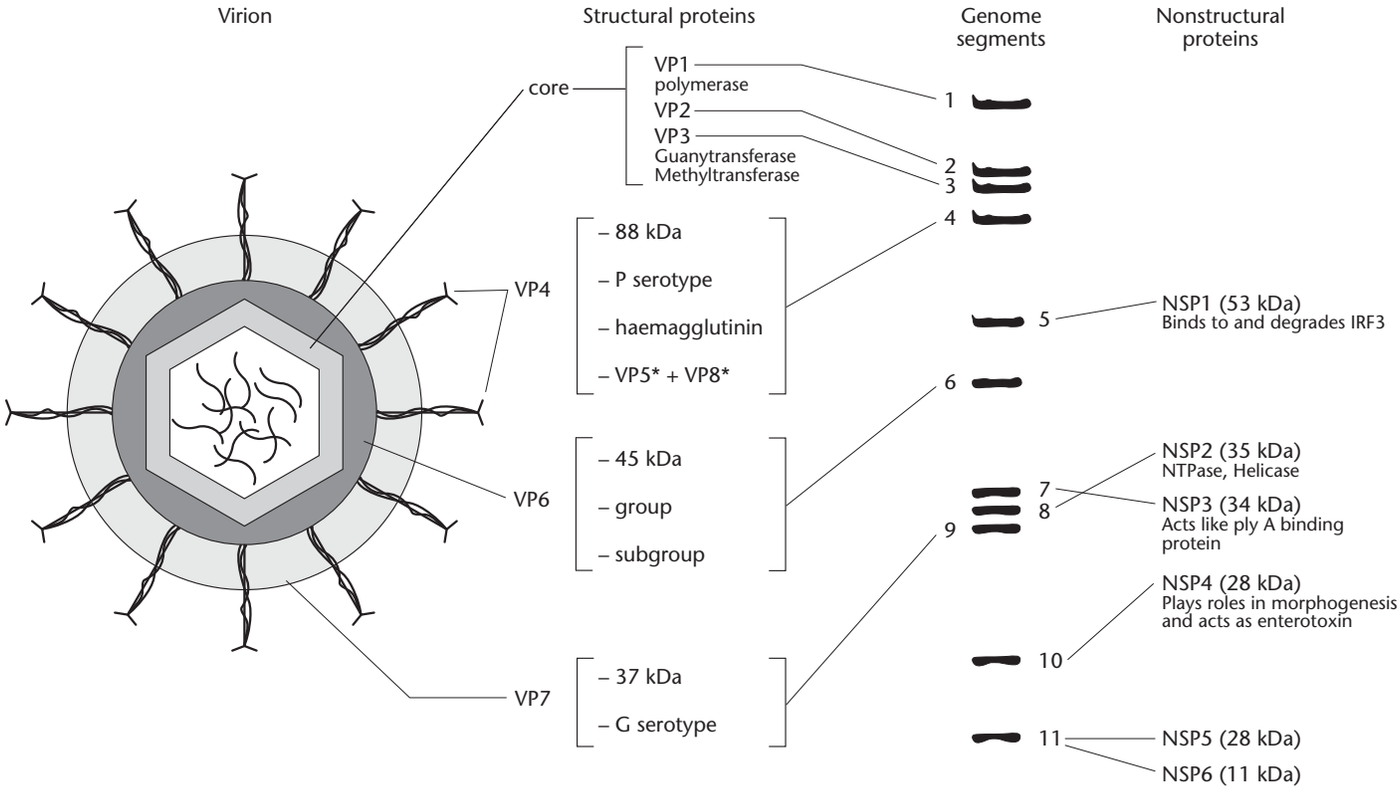

Figure 45.5 A schematic diagram showing the relationships between the structure of the rotavirus virion and the genomic double-stranded RNA segments. IRF3, interferon regulatory factor 3; NTPase, nucleotide triphosphatase.

enterocytes exceeds the rate of growth of immature enterocytes that are regenerated from the stem cells in the crypt, causing villous blunting and thus malabsorption. Infection resolves both as the virus runs out of susceptible mature enterocytes and an immune response is generated. Generally speaking, it is on only the first two or three occasions that disease occurs. However, it is now increasingly recognized that otherwise healthy adults can have rotavirus diarrhoea and elderly people appear to become more susceptible as their immunity wanes.[31,32] Recently, rotavirus antigen has been detected in the blood of immunocompetent infants as well as in experimentally infected animals.[33] The clinical significance of this finding is being investigated.

Immunity

In general, one or more episodes of rotavirus infection confers protection against subsequent severe rotavirus diarrhoea but not against asymptomatic reinfection or mild to moderate diarrhoea. In a cohort study in Mexico, children who had experienced one, two or three episodes of rotavirus diarrhoea had adjusted relative risks of experiencing a further attack of rotavirus diarrhoea of 0.23, 0.17 and 0.08, respectively, but of asymptomatic rotavirus infection of 0.62, 0.40 and 0.34, respectively.[34] Infection with one serotype provides serotype-specific (homotypic) protection, and repeated infections lead to partial cross-serotype (heterotypic) protection. Thus, serotype matters but it does not seem to be the sole determinant in providing protective immunity. Cellular immunity appears to be important in resolution of rotavirus infection and appears to be cross-protective between the different G serotypes.[35]

Protection of neonates against rotavirus infection appears to be by both transplacentally acquired maternal antibody[36,37] and by antibodies and other factors in breast milk.[38] However, a study in Bangladesh showed that hospitalized children with rotavirus diarrhoea were more likely to be breast-fed than were children with diarrhoea due to other infectious agents.[39] Interestingly, rotavirus infection in neonates often results in asymptomatic infection, unless novel serotypes emerge, and rotavirus can circulate silently in neonatal units. Since such asymptomatic neonatal infections induce protection against subsequent severe rotavirus gastroenteritis,[40] the use of neonatal strains as vaccine candidates has been pursued. However, a recent study in India showed that neonatal infection with a G10P[11] strain that resembles a neonatal vaccine candidate did not confer protection against subsequent rotavirus infection or diarrhoea of any severity.[41]

Clinical features

The outcome of rotavirus infection varies from asymptomatic, through mild short-lived watery diarrhoea, to an overwhelming gastroenteritis with dehydration leading to death. Severe disease and death are more common in children who are already malnourished or have measles. The onset of symptoms is abrupt after a short incubation period of 1–2 days. Fever, vomiting and watery

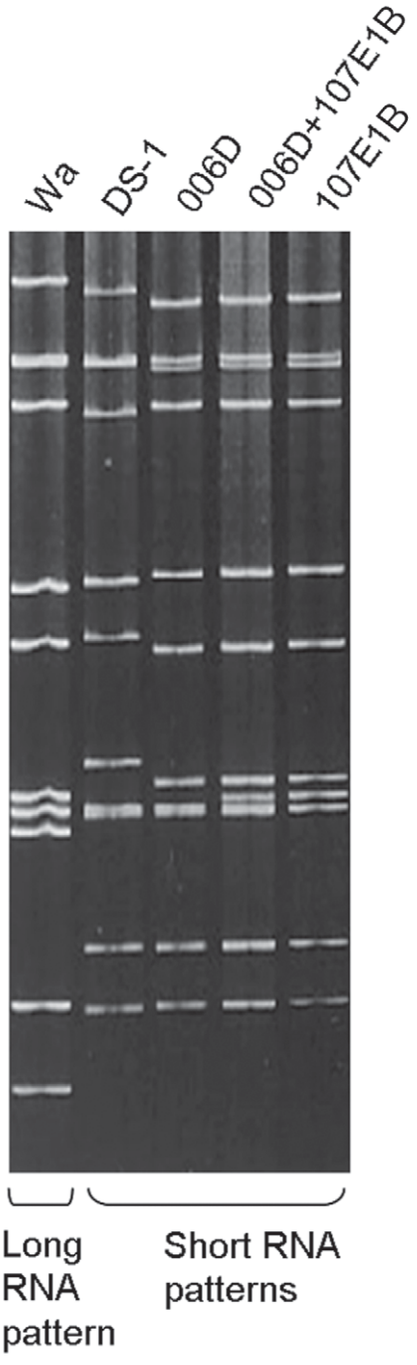

Figure 45.6 Separation of rotavirus genomic RNA into 11 bands by polyacrylamide gel electrophoresis. Two RNA patterns, long and short, are represented by prototype strains Wa and DS-1, respectively. Strains 006 and 107E1B have similar but distinct RNA electropherotypes. The differences in migration of segments 7, 8, and 9 are clearly demonstrated by co-electrophoresis in which RNAs from both 006 and 107E1B were loaded on the same lane. (Adapted from Nakagomi, et al.[17])

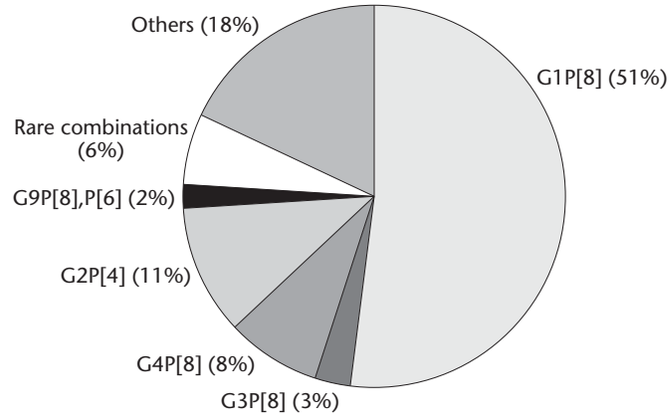

Figure 45.7 Relative frequencies of rotavirus genotypes detected globally among human rotaviruses over the period 1994–2003. (Adapted from Gentsch et al.[19])

diarrhoea are seen in the majority of infected children and last for 2–6 days. Rotavirus diarrhoea tends to be more severe than that due to other common enteropathogens[42] but co-infection with another pathogen does not increase disease severity.[43] Respiratory signs are often found during rotavirus gastroenteritis but its aetiological association with rotavirus infection is not clear. Extraintestinal manifestations during rotavirus gastroenteritis, including encephalopathies, have captured much attention since it was recently shown that rotavirus causes viraemia.[33,44] It is not possible to distinguish rotavirus gastroenteritis from other viral causes of non-inflammatory diarrhoea solely on clinical grounds. The stools are usually pale and watery or loose, and are seldom blood-stained. In hospitalized patients, the duration of diarrhoea is from 2 to 23 days, with a median of 6 days.[45] Patients continue to excrete virus for extended periods of time[25,46] and may thus be a reservoir for infecting others. The cause of death is dehydration, which can be hypo- or hypernatraemic and is often associated with metabolic acidosis.

Diagnosis

Rotavirus can be detected in stool specimens by a number of techniques, including electron microscopy, PAGE, antigen detection assays, RT-PCR and virus isolation. Electron microscopy is still a valuable diagnostic tool since it is a catch-all technique that will also detect other potential viral enteropathogens. PAGE is also a convenient diagnostic tool for the detection of rotavirus RNA extracted directly from stool specimens (Figure 45.6). The assay also allows detection of non-group A rotaviruses which fail to react in most antigen detection assays. This technique is relatively simple with good specificity (100%) and sensitivity (80–90%), and can be performed in tropical countries relatively cheaply.[47] It has an added advantage of providing epidemiological information because the electrophoretic migration pattern of the 11 segments of the double-stranded RNA genome is specific to each rotavirus strain.[18,48] Detection of viral genome by RT-PCR is a research tool which provides information on the G and P genotypes of the circulating strains[49–51] and the duration of viral shedding in stool.[25,46]

Antigen detection tests are currently the most widely used in diagnostic laboratories and include enzyme-linked immunosorbent assay (ELISA), latex particle agglutination assay, and immunochromatography.[52] The sensitivity and specificity of these tests are generally high (90–95%) but they are only designed to detect group A rotaviruses.

Group A and group C rotaviruses can be isolated in cell culture but viral culture is limited to research purposes. Similarly, antibody detection can be used for establishing a diagnosis but is not often employed.

Management

The mainstay of management consists of assessment of dehydration and replacement of lost fluid by oral rehydration with fluids of specified electrolyte and glucose composition. Intravenous rehydration therapy is indicated for patients with severe dehydration, shock or reduced levels of consciousness. Human or bovine colostrum and hyperimmune human serum immunoglobulin have been used to manage chronic rotavirus infection in immunocompromised children. Administration of probiotics such as *Lactobacillus casei* GG also appears beneficial. Recently, the antiprotozoal drug nitazoxanide was shown to decrease the median duration of rotavirus gastroenteritis by 44 hours in a randomized double-blind placebo-controlled trial in Egyptian children.[53] How this agent would be used in children in developing countries is unclear.

Prevention and control

Since virtually all children will have experienced rotavirus infection by the age of 3–5 years in both developing and developed countries, it is clear that the high standards of hygiene and sanitation practised in developed countries are not sufficient to prevent the spread of rotavirus infection within the community. Thus, prophylaxis of severe rotavirus gastroenteritis by vaccines remains as the only practical preventive measure.[4,54] The first licensed rotavirus vaccine, a rhesus monkey rotavirus-based tetravalent human reassortant vaccine (RotaShield®), was withdrawn after this live, oral vaccine was associated with the development of intestinal intussusception in approximately 1 : 10 000 vaccine recipients in the USA. Two new rotavirus vaccines, Rotarix® (GlaxoSmithKline Biologicals) and Rotateq® (Merck & Co.), have recently completed phase III clinical trials, each involving more than 60 000 infants. Both vaccines were found to be safe when given to infants under 3 months of age and were >85% efficacious in preventing severe gastroenteritis due to rotavirus.[55,56] Rotarix® is a monovalent human rotavirus vaccine of serotype G1P1A[8], whereas Rotateq® is a pentavalent bovine–human reassortant vaccine comprising types G1, G2, G3, G4 and P[8]. Updated disease burden estimates and economic justification will be needed wherever vaccine introduction is considered. Further confirmation of the safety profile of either vaccine will depend on post-licensure evaluation. Assessment of the ability of each vaccine to provide protection against an increasingly diverse population of rotavirus strains will require continuous global strain surveillance. Rotavirus does not produce more severe disease in HIV-infected infants,[57] so use of live-reassortant rotaviruses in populations with a high prevalence of HIV should not pose a risk.

ENTERIC ADENOVIRUSES

While adenoviruses have long been established as the cause of some respiratory and systemic infectious diseases and can be recovered in cell culture from stool specimens, they were not considered to be a causative agent of infantile diarrhoea until they were seen in large numbers in stool specimens. These adenoviruses were not cultivatable in cells used for the more conventional adenoviruses, and were thus called 'enteric' or 'fastidious' adenoviruses. They are now readily grown in 293 cells and are classified as serotypes 40 and 41 in group F adenoviruses.

Adenoviruses are unenveloped DNA viruses with an icosahedral capsid measuring 70–75 nm in diameter (Figure 45.8). Their genomes are double-stranded linear DNA of 33–45 kilobase pairs. Family Adenoviridae has now been divided into the genus *Mastadenovirus* (mammalian adenoviruses, to which human adenoviruses belong) and the genus *Aviadenovirus* (adenoviruses of birds). Human adenoviruses are further divided into six subgenera (A–F) and 51 serotypes.

Enteric adenoviruses account for approximately 5% of cases of infantile diarrhoea, occurring most often in children under 2 years of age.[58] There is no apparent seasonality to infection. Enteric adenoviruses are spread from person to person by the faecal–oral route. Neither food-borne nor water-borne spread has been described.

The clinical features of enteric adenovirus gastroenteritis do not differ greatly from those of rotavirus but the duration of diarrhoea tends to be longer in adenovirus infection than in rotavirus infection.[59,60] Other than gastroenteritis, adenovirus is implicated as a cause of idiopathic intussusception in infants.[61,62] These adenoviruses are of serotypes 1, 2, 3 and 5, and rarely of 40 or 41 (enteric adenoviruses).

The diagnosis of adenovirus infection is by visualization of characteristic virions in stool specimens under the electron microscope; demonstration of adenovirus antigens in stool by ELISA,

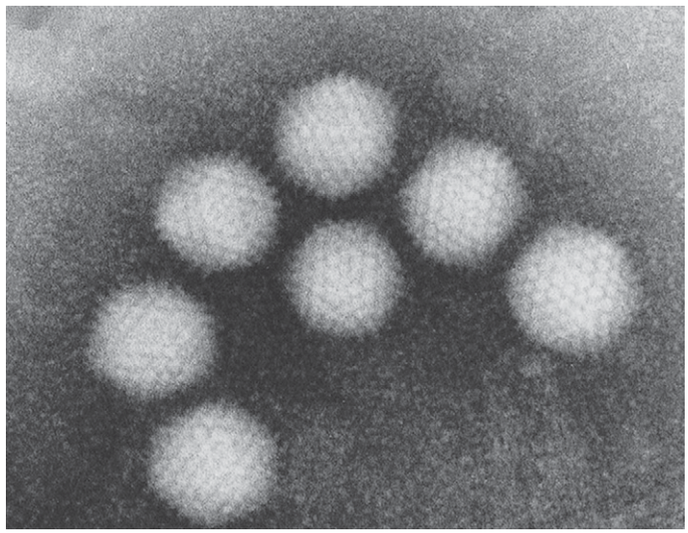

Figure 45.8 Negative-stain electron micrograph of enteric adenovirus. (×200 000.)

latex agglutination assay or immunochromatography; or detection of the genome by DNA hybridization (mostly in tissues) and PCR.

Treatment of adenovirus diarrhoea is by managing dehydration. There is neither a specific therapeutic intervention nor a vaccine.

ASTROVIRUS

Astrovirus was first described in 1975 and is now established as an important cause of gastroenteritis in children and adults. The family Astroviridae can infect a variety of animal species, including humans. Astrovirus is an unenveloped virus measuring 28–30 nm in diameter. Under the electron microscope it has a characteristic star shape 'stamped' on its surface, a five- or six-pointed star with an electron-dense centre (*astron* is Greek for a star) (Figure 45.9). It has a positive-sense single-stranded RNA genome approximately 7 kb in length which encodes an RNA polymerase (ORF1a), a serine protease (ORF1b) and three capsid proteins (ORF2).

There are at least eight serotypes (serotypes 1–8) of human astroviruses and serotype 1 is the most frequently detected.[63] However, other serotypes can be responsible for outbreaks of food-borne infections and there appears to be a greater diversity of serotypes in developing countries.[64] The importance of astrovirus gastroenteritis has only recently been recognized with the development of improved diagnostic tests such as ELISA. Astrovirus infections predominate in young children aged between 4–5 months and 4 years, and account for between 2% and 10% of cases of diarrhoea in children. The disease tends to be milder and more frequently encountered in community-based studies.[65] One such study in Mexico estimated the incidence rate of astrovirus gastroenteritis to be 0.1 episodes per child per year.[66] Sero-epidemiological studies have demonstrated that more than 90% of children in the USA will have experienced astrovirus infections by the age of 6–9 years.[67] Astrovirus has been detected in all countries where sufficiently sensitive detection methods have been used, including Malawi,[68] Mexico,[66] South Africa,[69] and Egypt.[70] In temperate countries it shows a similar seasonal distribution to rotavirus but peaks a month earlier.

Astrovirus is transmitted faeco-orally either directly or by ingestion of food. Astrovirus infects the upper small intestine but the mechanism of diarrhoea is not known. The features of the illness are similar to those of rotavirus but may be milder and its duration is 4–5 days on average. However, in Bangladesh, astrovirus was found to be associated with prolonged diarrhoea.[64]

Diagnosis used to be solely by electron microscopy, but this is now being replaced by more sensitive and easy-to-perform ELISA or latex agglutination assays, or by detection of the genome by RT-PCR. Treatment is by managing dehydration. There is no vaccine available and little is known of immunity to infection other than that children with immunodeficiency excrete the virus for long periods.[71]

CALICIVIRUSES

Norovirus and *Sapovirus* are two different genera of viruses that belong to the family Caliciviridae and are major causative agents of acute gastroenteritis in children and adults. The Norwalk agent, the prototype of *Norovirus*, was first identified by immune electron microscopy as 27 nm virus particles with a feathery-ragged outline (Figure 45.10) in stool specimens of volunteers who were challenged with the clinical specimens collected during a gastroenteritis outbreak in Norwalk, Ohio, USA.[72] In addition, stool examination by negative-stain electron microscopy has demonstrated the presence of 'typical' calicivirus-like particles (Figure 45.11), now classified as *Sapovirus* (the prototype was found in Sapporo, Japan), in the stool specimens of children. These exhibit the 'classical' distinct cup-shaped depressions (*calyx* is Greek for a cup) on the surface of the virion.[73,74] Molecular cloning of these viruses has confirmed that both noroviruses and sapoviruses are members of Caliciviridae but each constitutes a distinct genus within the family.[75]

Norovirus

Norovirus has an unenveloped virion with icosahedral symmetry, measuring 27–30 nm in diameter. Its genome is positive-sense, single-stranded RNA, approximately 7 kb in length, with a stretch of poly A sequence at its 3′ terminus. The genome contains three

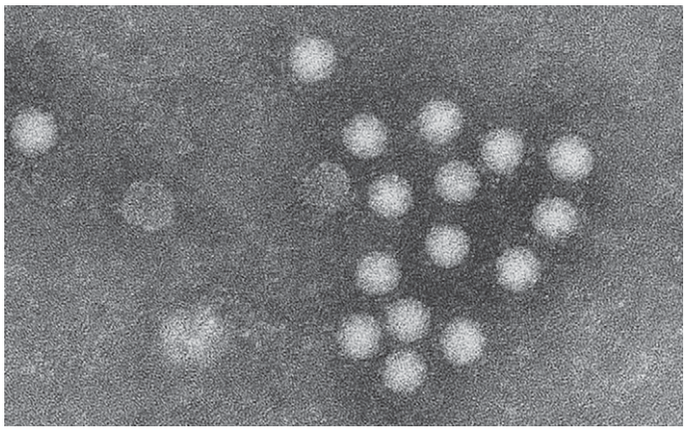

Figure 45.9 Negative-stain electron micrograph of enteric astrovirus. (×200 000.)

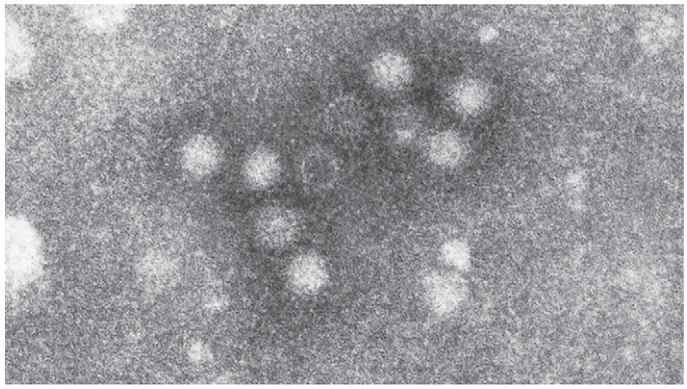

Figure 45.10 Negative-stain electron micrograph of norovirus with a feathery-ragged outline. (×200 000.)

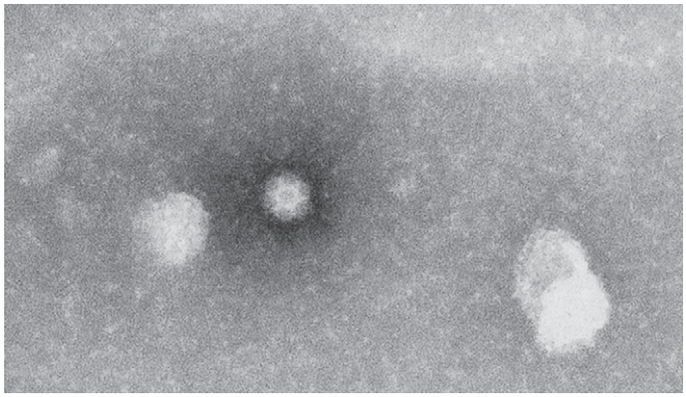

Figure 45.11 Negative-stain electron micrograph of a sapovirus with the classical 'Star of David' morphology. (×200 000.)

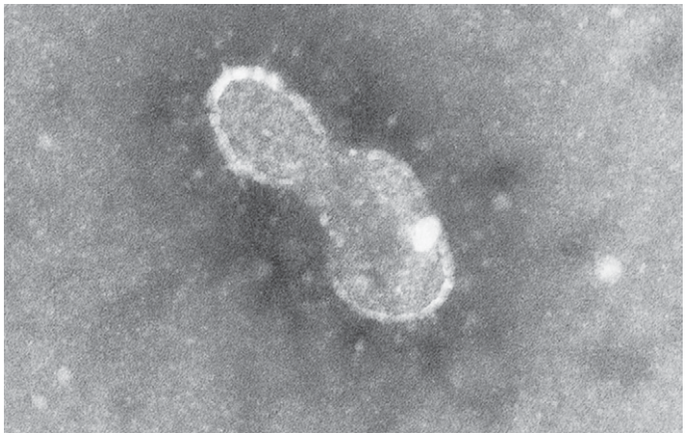

Figure 45.12 Negative-stain electron micrograph of an enteric coronavirus. (×200 000.)

ORFs, of which ORF2 encodes a single polypeptide of 59 kDa on which the antigenicity of a virus strain is expressed. Since neither animal model nor cell culture systems are available to test the infectivity of norovirus other than volunteer challenge studies, serotypes of norovirus have not been established. The genome of norovirus exhibits a great diversity and there are multiple genotypes that are distributed into four genogroups (GI–GIV).[76]

Norovirus is spread faeco-orally, and causes an illness with an abrupt onset of vomiting, diarrhoea and abdominal pain after a short incubation period of 1–2 days. While transmission via the respiratory route has not been established, it has been suggested from epidemiological observations that aerosolized saliva or vomitus can be the source of infection.[77] The illness is generally mild and fever rarely exceeds 38 °C. Recovery follows within 1–3 days, but the excretion of norovirus into stool lasts longer, sometimes up to 2 weeks. Approximately half of those infected with norovirus remain asymptomatically infected. In temperate countries, norovirus gastroenteritis tends to show a winter seasonality. Infection can occur as point-source food-borne outbreaks or sporadically.

Norovirus was initially thought solely to be the cause of epidemic gastroenteritis limited to older children and adults, but it is now known that norovirus is also a major cause of infantile diarrhoea. In a study in Finnish children, norovirus was responsible for 20% of cases of gastroenteritis. In comparison, sapovirus was detected in 9%, astrovirus in 10%, enteric adenovirus in 6%, and rotavirus in 31% of the cases.[78]

Electron microscopy is relatively insensitive except in the first days of the illness and the definitive diagnosis needs to be made based on either antigen detection or identification of the norovirus genome by RT-PCR.[79] Recently, commercial antigen detection kits have been developed, which are less sensitive than genomic detection by RT-PCR but the specificity is close to 100%.

Following infection, patients produce serum and faecal antibody to viral capsid proteins but their role in protective immunity is not fully defined. Recently, considerable progress has been made in understanding the relationships between norovirus and tissue antigens, including the ABO blood groups.[80] Norovirus appears to use secreted blood group (H) antigens expressed on the mucosal surface of the enterocytes as viral receptors; thus, non-secretors, in whom such antigens are not expressed on the intestinal mucosa, are resistant to norovirus infection.[81]

There is no specific therapy nor are vaccines available.

Sapovirus

Sapovirus has an unenveloped virion with icosahedral symmetry, measuring 30–35 nm in diameter. Negative-stain electron microscopy reveals characteristic particle morphology with cup-like depressions, often described as the 'Star of David' (Figure 45.11). Its genome is positive-sense, single-stranded RNA of approximately 7 kb in length with a stretch of poly A sequence at its 3′ terminus. Unlike norovirus, sapovirus encodes the capsid protein contiguous with the large non-structural polyprotein (ORF1). The junction that corresponds to ORF1 and ORF2 of norovirus consists of a one- or four-nucleotide overlap between the stop codon of ORF1 and the first AUG codon of ORF2. This creates a −1 frameshift. The 3′ end of ORF1 encodes a single polypeptide of 62 kDa.

Illness due to sapovirus tends to predominate in young children, and virtually all children appear to have experienced infection by sapovirus by the age of 5 years. In temperate countries, sapovirus gastroenteritis occurs more frequently in winter. Sapovirus accounts for approximately 5% of cases of infantile diarrhoea, the detection rates being similar to those of adenovirus and astrovirus. Sapovirus rarely causes outbreaks of food-borne gastroenteritis.

Sapovirus spreads faeco-orally and infects, and causes predominantly diarrhoea in infants and young children. Protective immunity appears to follow infection, since, unlike with norovirus, adults rarely get sapovirus gastroenteritis.

While a typical calicivirus-like morphology under the electron microscope strongly suggests the presence of sapovirus, the definitive diagnosis needs to be made based on either antigen detection or identification of the sapovirus genome by RT-PCR.[79]

Treatment is by management of dehydration. There is neither specific antiviral chemotherapy nor a vaccine available.

OTHER VIRUSES

A number of other viruses, including coronavirus[82] (Figure 45.12), torovirus,[83] picobirnavirus[84,85] and pestivirus,[86] have been detected in stool specimens of patients with acute gastroenteritis, but their significance as aetiological agents of diarrhoea remains to be established.

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