Rotaviruses were first described in 1963 when they were observed during electron microscopy of faecal samples from monkeys and mice. Some 10 years later, similar viruses with the characteristic spherical structure resembling the spokes of a wheel were observed in faecal samples from children with diarrhoea (Fig. 1).

The capsid, a protein coat that surrounds the genome, has three layers each constructed from a different viral protein (VP). The inner and middle layers, constructed from VP2 and VP6, respectively, are perforated by channels. The middle layer contains the ‘spokes’ of the wheel and is a major constituent of the virion. The outer layer is constructed from another protein, VP7, which is a glycoprotein. Three other proteins are found in the virion, VP1 and VP3 in the core, and VP4 which forms 60 spikes on the surface (Fig. 2). Both VP7 and VP4 have a role in protective immunity.

The proteins are numbered in order of their size, the largest being towards the centre of the virion. Associated with the genome within the inner capsid layer are copies of VP1 and VP3 which are enzymes (Fig. 3).

The genome comprises 11 double stranded RNA segments. Each segment encodes one protein except one segment which encodes two proteins. Thus in all 12 proteins are encoded—six structural and six non-structural proteins.

The two structural proteins that comprise the outer viral capsid are the neutralization antigens VP7 (or G for glycoprotein) and VP4 (or P for protease sensitive). They define the serotypes [1]. There are 23P and 15G serotypes. Five G–P combinations G1P8, G2P4, G3P8, G4P8 and G9P8 constitute 90% of human rotavirus strains worldwide. G1P8 strains are most prevalent but large regional differences exist and strain types can change with time [2]. This has implication in the development of vaccine.

Change in serotypes occurs because of:

(i) Point mutation (or ‘drift’).
(ii) Genomic reassortment caused by exchange of RNA segments between strains (or ‘shift’). Overall most reassortment occurs in the genes encoding VP7 and VP4.
(iii) Duplication and deletion of sequences within a segment.
(iv) Introduction of animal rotaviruses into the human population.

Reassortment and crossing of species barrier results in a large number of G types besides just the G1 and G4 in some regions. Thus serotypes G5, G8 and G9 have been described as important serotypes encountered in Brazil, Malawi, India and Bangladesh, respectively [2, 3].

The virion utilizes sites on the spike proteins initially to bind to the host cell, followed by sites on the glycoprotein (VP7) on the capsid surface. On entry into the host cell the virus elaborates potent exotoxin that can cause diarrhoea, and then goes on to destroy the cell causing blunting of villi, extensive damage and shedding of large quantities of virus. The faeces of the infected individual may contain up to $10^9$–$10^{10}$ per gram. A small infectious dose ($<100$ virus particles) is needed which facilitates transmission.

In developing countries rotavirus diarrhoea is the third most common cause of death in infants with an estimated 5% of all deaths in children <5 years old [4, 5]. First infections after the neonatal period are generally symptomatic, some 1–2% developing severe or moderately severe disease. Repeat disease is uncommon after the second infection.

Antibody response mounted by the host is targeted at two proteins of the outer capsid namely VP7 (a G protein) and VP4 (a P protein). Vaccine development has aimed at promoting antibody response to these capsid proteins of the commonly occurring strains. Animal strains from monkeys (rhesus RRV) [6], cows (WC 3) [7, 8] and lambs [9], that are not infective for humans have been cultured with human strains to produce reassortants. Those reassortants selected for vaccine production contain 10 genes from animal strains to maintain non-infectivity and one gene-encoding proteins of outer capsid common to the circulating serotype.

A candidate oral vaccine derived from bovine strain of rotavirus (RIT4237) underwent a trial in 1983 with disappointing results [10]. The efficacy of monovalent vaccine was improved by reassortants strains that were non-infective tetravalent rhesus strains as well as individual genes encoding outer capsid proteins of common human strains [11]. The vaccine (Rotashield, Wyeth-Lederle, Pearl River, NY, USA) was found highly effective in clinical trials. But an unexpected complication, intussusception, was reported to occur in the first two weeks after administration of the first dose resulting in the vaccine being withdrawn [12].
Two next generation vaccines based on different principles have recently undergone clinical trials. Rotarix (GlaxoSmithKline, Belgium) is derived from an individual human strain that replicates well in the gut and is shed in the stools. The strain was at first attenuated by passaging 43 times, then cloned and renamed RIX4414 and further passaged [13]. The vaccine is in the form of a lyophilized powder which must be reconstituted with 1 ml of citrate bicarbonate buffer and is administered orally. Following initial trials in Finland that showed safety, immunogenicity and efficacy [14], the vaccine was tested in middle-income countries of Latin America (Brazil, Mexico and Venezuela) [15] and in Singapore [16]. The vaccine showed an efficacy of 70–85% against rotavirus diarrhoea and 85–93% against severe disease. A large multicentre trial that enrolled 63 000 infants in 12 countries of Latin America and Finland was next undertaken. The effectiveness of the vaccine was 85% against severe rotavirus diarrhoea, and reduced admission for diarrhoea of any cause by 41%. Six vaccinated subjects and seven placebo administered ones developed intussusception during 1 month of observation confirming lack of causal association.

Rota Teq (Merck) is prepared from a bovine strain of rotavirus (WC3) that had shown variable effectiveness in clinical trials in USA, China and Africa. To improve its effectiveness reassortant strains were prepared containing 10 genes from the parent bovine strain and an individual capsid gene from the most common human serotypes. Five single gene reassortants each containing a gene for a capsid protein for human serotypes G1–G4 and PIA were combined in a pentavalent vaccine [17]. The vaccine is formulated as liquid containing $12 \times 10^7$ infectious units per dose, and is administered in three oral doses commencing at 6 weeks of age. Efficacy of 96% against admission for rotavirus diarrhoea was demonstrable. The vaccine also protects against a range of rotavirus serotypes in circulation [18].

**Other Candidate Vaccines**

Regional diversity of rotaviruses requires that vaccines evolved from locally prevalent genotypes are likely to be more efficient. Several vaccines based on this principle are at present in experimental stage. In China, a strain of rotavirus obtained from lamb and grown in calf kidney cells has been the starting point of a vaccine licensed in that country. In India, two candidate rotavirus strains obtained during nosocomial outbreaks in asymptomatic newborns, strain 116E and 1321 are likely source of a vaccine. Both strains do not cause disease in infants. They are natural human bovine reassortants that grow well in the infant gut, and can resist neutralization by maternally derived antibodies [19, 20]. Another naturally occurring asymptomatic neonatal rotavirus

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**Fig. 1.** Electron microscope appearance of rotavirus.

**Fig. 2.** (a) On electron microscopy and (b) schematic representation.
strain in Australia, RV3, is also under consideration as a potential source of vaccine [21].

Since vaccines are licensed on the basis of safety, immunogenicity and efficacy candidate vaccines will need to be tested in clinical trials in a number of low-income countries to check their effectiveness where exclusive breastfeeding is the norm, malnutrition is prevalent and frequent transmission of enteric microorganisms occur in a generally unhygienic environment. A robust vaccine to protect infants in such an environment is the overall aim.

G. J. Ebrahim
<ebrahim@waitrose.com>

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